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Yong-Ku Kim *Editor*

# Neuroinflammation, Gut-Brain Axis and Immunity in Neuropsychiatric Disorders

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# Preface

A large body of evidence indicates inflammation may play a role in the pathophysiological mechanisms underlying mental illnesses such as depression, bipolar disorder, and schizophrenia and neurodegenerative disorders. Classic anti-inflammatory drugs such as corticosteroids and nonsteroidal anti-inflammatory drugs (such as selective cyclooxygenase-2 inhibitors) have shown consistent beneficial effects in patients with mood disorders and schizophrenia. Moreover, supporting evidence indicates that psychotropic medications affect the production of inflammatory mediators in both animals and humans.

Despite significant support for the "inflammation hypothesis" of mental disorders, many unanswered questions and controversies remain. For example, (a) many studies have reported that inflammatory mediator profiles are not altered among mentally ill patients; (b) the majority of human studies have examined peripheral tissues (especially blood), but brain tissue is more pertinent to the study of psychiatric illnesses; (c) it is not clear whether the therapeutic efficacy and toxicity of psychotropic drugs are influenced by inflammation; and (d) some studies reported that anti-inflammatory compounds were not effective as treatments for mental disorders.

There is increasing evidence that glial cells perform many important roles in various brain functions. A greater understanding of the interaction between neurons and glia may shed new light on clarifying many unknown aspects including the mind-brain gap and conscious-unconscious relationships. It is well known that central nervous system (CNS) inflammation and immune activation play a major role in the pathophysiology of neurodegenerative diseases. Although the blood-brain barrier is able to protect the CNS from immune activation, it becomes more permeable during inflammation, which renders the brain vulnerable to infections. A better understanding of the interaction between inflammatory mediators, such as cytokines, and the activated immune response, including astrocytes and microglia, is critical for the development of new therapeutic strategies for neurodegenerative diseases.

The gut-microbiota-brain axis is an area of active research with respect to neuropsychiatric disorders and their pathophysiological mechanisms. Bidirectional interactions between microorganisms and the brain affect various CNS activities (such as the stress response, behavior, and mood) through immune and

neuroendocrine system pathways. The gut microbiota are thought to directly or indirectly influence neuropsychiatric illness. Various neuropsychiatric disorders (including autism, depression, anxiety, and schizophrenia) are associated with or modulated by variations in the microbiome, microbial substrates, and by exogenous prebiotics, antibiotics, and probiotics. The microbiota-gut-brain axis may provide novel targets for prevention and treatment of neuropsychiatric disorders.

This book reviews the latest research addressing the relationships between cytokines, glia, and neurons in the pathophysiology of neuropsychiatric disorders and examines the mechanisms of action of the drugs used for treatment of these disorders. Evidence indicating inflammation-induced production of toxic metabolites from the tryptophan pathway plays a role in a wide range of neuropsychiatric disorders, including depression, bipolar disorder, and Alzheimer's disease, is provided. In presenting a review of the state of the science with regard to the interactions between cytokines, glia, and neurons, the book will help to pave the way for the development of novel targets for the prevention and treatment of neuropsychiatric disorders.

I sincerely thank all of the authors for their valuable time that was spent preparing manuscripts.

Ansan, Republic of Korea Yong-Ku Kim

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# Part I

# Rethinking and Paradigm Shift

# Neuron-Microglia Crosstalk in Neuropsychiatric Disorders

1

Sang Won Jeon and Yong-Ku Kim

**Abstract**

Numerous studies have investigated the causes and mechanisms of psychiatric disorders through postmortem examination of patients with a history of a schizophrenia, mood disorder, or neurocognitive disorder. In addition, the search for specific mechanism-based treatments for psychiatric disorders has been intensified through the use of transgenic animal models involving specific genes tightly associated with psychiatric disorders. As a result, many studies with patients or animal models have reported a close association of neuroglia with major psychiatric disorders. Recently, research has focused on the associations between microglia and major psychiatric disorders and on the role of the immune response and abnormal microglia in the onset and symptoms of psychiatric disorders, in particular. Postmortem studies of brain tissue and animal models recapitulating human mental disorders have also confirmed association between psychiatric disorders and quantitative, structural, or functional abnormalities of neuron-microglia crosstalk. This review aims to describe the relationships between microglia and major psychiatric disorders and to specifically examine studies of gene expression and function of microglia in depression, schizophrenia, and Alzheimer's disease.

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**Keywords**

Microglia · Neuroglia · Neuron · Depression · Schizophrenia · Alzheimer's disease

## 1.1 Introduction

Neuroglia is a generic term that defines nonneuronal cells in the nervous system. Glia account for 90% of the cells of the central nervous system (CNS) and consist of oligodendroglia, astroglia, and microglia cells [1]. The definition of neuroglia has recently been extended to include nerve/glial antigen 2 (NG2) cells, a type of progenitor cell distributed throughout the cerebrum with unique differentiation patterns in each brain area [2]. The basic function of neuroglia is to defend the central nervous system and maintain homeostasis. As with neurons, most neuroglia cells are derived from neuroepithelial cells; as such, they may resemble neurons in terms of structural and molecular characteristics, although unlike neurons, neuroglia lack axons and dendrites and have highly heterogeneous cellular morphology as a result of their optimization for various homeostatic functions [3]. In addition, while most mature neurons (except those in certain brain areas) lack the ability to proliferate by mitosis, neuroglia are able to proliferate in a suitable environment [4].

Neuroglia support neurons through glucose shuttling and phosphorylation to control synaptic plasticity and may also produce cytokines. The loss of neuroglia has been consistently reported in postmortem studies of individuals with psychiatric disorders [5]; additionally, the distribution of cytokine receptors throughout the hypothalamus, hippocampus, locus ceruleus, and prefrontal cortex also implies the role of neuroglia, especially the immune responses of microglia, in the pathogenesis of a variety of psychiatry disorders [6]. This review aims to describe the associations between microglia and psychiatric disorders and, specifically, the role of abnormal immune responses and microglial functions in the onset and symptoms of major psychiatric disorders.

## 1.2 Physiological Roles of Microglia

Microglia account for approximately 5−15% of brain cells and are considered the resident macrophages of the central nervous system (CNS). Given that microglia originate from hematopoietic precursor cells, they act as the immune cells of the CNS and assist in interactions between the immune system and glutamate neurotransmission [7]. In CNS disease, including neurodegenerative diseases, stroke, traumatic injury, or brain tumors, microglia migrate to and surround the damaged or dead cells to remove the cellular debris [8]. Microglia engage in apoptosis, proliferation, and differentiation of neurons during neurogenesis with crucial roles in the neurogenesis of mature cells [9, 10]. The main physiological functions of

microglia may be categorized as proliferation, morphological transformation, motility and migration, intercellular communication, phagocytosis, and proteostasis [11].

Microglia are capable of regulating the extracellular milieu and inflammatory responses through the expression of K+ channels and release of various proteins such as cytokines, similar to the activities of astroglia [12]. In addition, microglia may contribute to neuroregeneration through the secretion of neurotrophic factors such as the brain-derived neurotrophic factor (BDNF) and nerve growth factor (NGF) [13]. Microglial secretion of BDNF is known to promote synaptic plasticity, as well as learning and memory, by increasing the expression of the glutamate receptors GluN2B and VGluT1 at the synapse [14]. Microglia create an appropriate environment for normal brain function by acting as immune cells that support and interact with neurons. The general hypothesis is that microglia remain quiescent under normal conditions and become activated upon injury or disease. However, microglia are morphologically dynamic, continuously extending their branch-like protrusions into the extracellular space and migrating through the CNS [15], which enables them to monitor the cerebral parenchyma and maintain tissue homeostasis, with rapid responses to brain injury or damage [16].

## 1.3 Neuron-Microglia Crosstalk in Depression

In cases of major depressive disorders, the number of astroglial cells in the cerebral cortex and limbic system of the patients is reduced. The reduction in the number of astroglial cells may lead to abnormal synaptic structure or glutamate function in neurons and an imbalance in energy metabolism, leading to dysfunctions in the cerebral cortex and limbic system [17]. In an animal model, when the number of astroglia cells in the cerebral cortex was specifically reduced, a state of depression appeared, indicating a close association of depressive disorders with the reduced number of astroglia [18]. White matter obtained from patients with major depressive disorder was found to have greatly increased thickness of the myelin sheath generated by oligodendroglia cells, in contrast to the findings in schizophrenia patients [18], while the prefrontal cortex showed an increase in the number of microglia cells [19]. In addition, in an animal model, activation of the nucleotide binding and oligomerization domain-like receptor family pyrin-domain-containing 3 (NLRP3) inflammasome in microglia cells of the prefrontal cortex was reported to increase interleukin-1 beta (IL-1β) production, which was suspected to be the causal mechanism of major depressive disorder [20]. In contrast, bipolar disorder was correlated with a reduction in the number of oligodendroglia cells in humans [21], while abnormal activation of the glycogen synthase kinase (GSK)-3 beta, as a transmission signal related to serotonins in microglia, promotes the onset of bipolar disorder [22].

The main cytokines released by microglia, T-helper 1 (Th1) lymphocytes and M1 macrophages, are the pro-inflammatory Th1-type cytokines, including IL-1β, IL-2, IL-6, tumor necrosis factor-α (TNF-α), and interferon-γ (IFN-γ) [23]. Pro-inflammatory cytokines activate cyclooxygenase-2 (COX-2), which induces prostaglandin E2

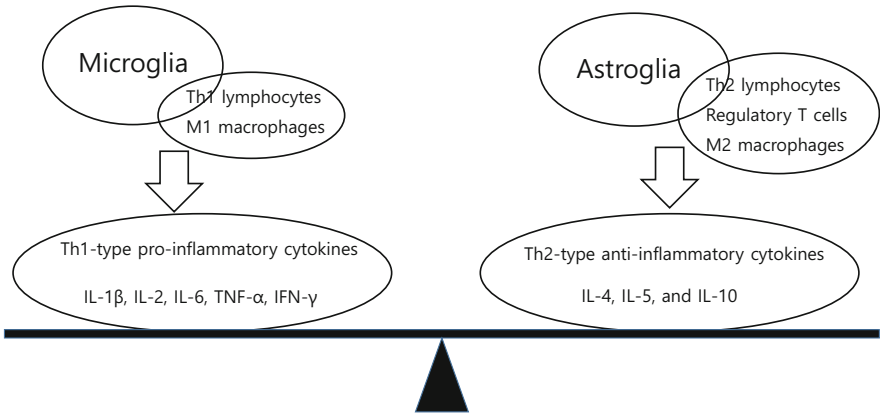


**Fig. 1.1** Th1-Th2 cytokine seesaw

(PGE2) expression, thus increasing the activity of inflammatory cells and promoting inflammation. In contrast, astroglia cells, Th2 lymphocytes, regulatory T cells (T regs), and M2 macrophages mainly release Th2-type anti-inflammatory cytokines, including IL-4, IL-5, and IL-10 [23]. The interaction of these cytokines is known as the "Th1-Th2 cytokine seesaw" (Fig. 1.1), whereby inflammation is determined according to the relative dominance between Th1 and T2 cytokines. Together, microglia, astroglia, T cells, and glutamate activities are key contributors to the inflammatory cytokine profile [24], and during chronic inflammation, an imbalance in cytokine profiles appears to lead to the onset of various mental disorders [25].

An imbalance in the Th1-Th2 cytokine production in the CNS affects the metabolism of tryptophan, a precursor of serotonin. Indoleamine-2,3-dioxygenase (IDO) secreted by microglia and astroglia is the rate-limiting enzyme in the metabolism of tryptophan to kynurenine and of serotonin to 5-hydroxyindoleacetic acid (5HTT). Kynurenine 3-monooxygenase (KMO) secreted by microglia is the rate-limiting enzyme in the transformation of kynurenine into 3-hydroxykynurenine, while tryptophan-2,3-dioxygenase (TDO) and kynurenine aminotransferase (KAT) secreted by astroglia are the rate-limiting enzymes in the transformation of tryptophan into kynurenine and kynurenine into kynurenic acid, respectively [26]. In the CNS, when Th1 cytokines are dominant over Th2 cytokines, neuroglia cells increase the secretion of IDO and KMO, with a consequent reduction in serotonin and increase in kynurenine. Thereafter, kynurenine is converted into quinolinic acid, which is an n-methyl-D-aspartate (NMDA) receptor agonist in microglia, to increase glutamate neurotransmission as well as intracellular calcium influx, which in turn reduces Th2 activity but promotes Th1 activity in astroglia cells, thus perpetuating inflammation [26, 27]. In addition, kynurenine is converted to kynurenic acid in astroglia cells, which is an NMDA receptor antagonist, which decreases glutamate neurotransmission (Fig. 1.2) [28].

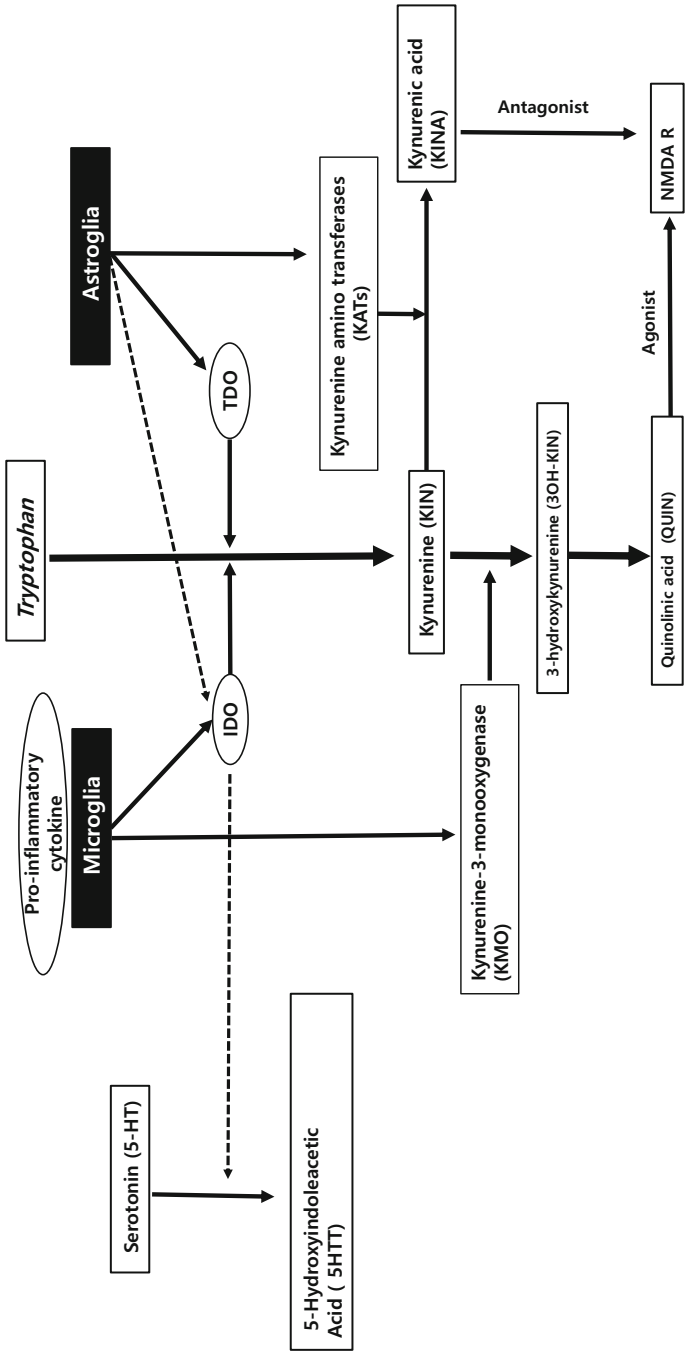


**Fig. 1.2** Neuron–microglia crosstalk in depression

These phenomena support the glutamate and neuroplasticity model of major depressive disorder. Notably, glutamate concentrations in the cerebral cortex are proportional to the severity of depressive symptoms. Given that ketamine, an NMDA receptor antagonist, exhibits highly potent antidepressive effects, this model may explain the pathophysiology of major depressive disorder [29]. It is thus likely that the role of microglia and cytokine imbalance in major depressive disorder will receive continued attention. In a meta-analysis including 29 previous studies that compared patients with major depressive disorder ($n = 822$) and healthy control individuals ($n = 726$), increased levels of soluble IL-2 receptor (sIL-2R), IL-6, and TNF-α were determined to be markers of major depressive disorder in the blood sample [30]. In a comparison of patients with major depressive disorder with ($n = 47$) or without ($n = 17$) a history of suicidal attempt and healthy controls ($n = 16$), increased TNF-α and IL-6 and reduced IL-2 were correlated with suicidal attempts [31]. The degree of alteration in cytokine profiles varied across the reviewed studies, presumably due to varying past histories and disease durations of participating patients. Among the various cytokines, IL-6 exhibited the most consistent increase and was thus considered a marker of the risk of suicide and treatment outcomes in relation to major depressive disorder [32]. However, as cytokines were often measured in peripheral blood samples, the CNS status of the study patients could not be confirmed due to potential confounding by a number of factors, such as physical disorders, leading to some uncertainty in the interpretation of the results. In addition, studies assessing cytokines in CNS tissue samples were often small-scale in vitro studies. As a consequence, recent studies have instead examined S100 calcium-binding protein B (S100B) as a novel marker of neuropsychiatric disorders.

The S100B peptide is secreted by neuroglia and is involved in the control of calcium homeostasis, as it acts on the receptor for advanced glycation end product (RAGE) in neurons and neuroglia [33]. Depending on the concentration, S100B may protect or damage neurons [33]. Because it can pass through the blood–brain barrier (BBB) and may be detected in the peripheral blood, S100B is also called the "C-reactive protein (CRP) of the brain" and serves as a clinical indicator of cerebral ischemia, hemorrhage, trauma, and neurodegenerative diseases [34]. Increased plasma concentrations of S100B have also been detected in mood disorders and were of notable significance in acute major depressive episodes and manic episodes and have been positively correlated with the severity of suicide [35]. Increased plasma S100B concentrations in schizophrenia patients were positively correlated with paranoia, negative symptoms, cognitive impairment, reduced therapeutic response, and disease duration [36]. Plasma S100B concentrations decreased in response to treatment with antidepressants and antipsychotic drugs [37], and the genetic polymorphisms of *S100B* and *RAGE* genes detected in patients with depressive disorder or schizophrenia indicated they may be associated with the primary etiology of the disease, rather than being secondary markers [38].

## 1.4 Neuron-Microglia Crosstalk in Schizophrenia

A key difference between schizophrenia and neurodegenerative diseases, in terms of pathology, is that the former exhibits a reduced volume of specific brain regions (frontal lobe, parietal lobe, temporal lobe, hippocampus, and parahippocampus) without a prominent loss of neurons [39]. Brain imaging and postmortem examinations of patients with schizophrenia and animal disease model studies showed various abnormal findings in the white matter, thus highlighting the role of structural abnormalities in neural connectivity or neural network connectivity [40]. These results have shifted the focus of research into the pathology of schizophrenia toward the role of neuroglia cells that are the primary component of white matter. While the number of studies reporting abnormal findings related to neuroglia in schizophrenia has increased, it is still unclear whether abnormal neuroglia are the primary cause or a secondary effect of schizophrenia [41].

Previous studies of patients with schizophrenia and animal models found associations between the onset of schizophrenia and increased numbers of astroglia cells in the amygdala and cerebral cortex [42]. In schizophrenia patients, increased numbers of astroglial cells in the cerebral cortex that express the retinoic acid-inducible gene 1 (RAI-1), which regulates the retinoic acid signaling pathway that plays a critical role in synaptic plasticity and learning, were determined to be a key determinant of schizophrenia symptoms and treatment responses [43]. Other factors that induce schizophrenia are a quantitative reduction of oligodendroglia cells, reduced expression of genes related to the myelin sheath of oligodendroglia cells, and abnormal differentiation of oligodendroglia cells during development [44]. Two genes that cause schizophrenia, disrupted-in-schizophrenia-1 (*DISC1*) and DISC1-binding zinc finger (*DBZ*), control differentiation of oligodendroglia cells, and mutations of these genes have the potential to cause schizophrenia by leading to dysfunctions in the differentiation of and myelin sheath formation by oligodendroglia cells [44].

One study also reported an increase in the density of activated microglia cells in the brain of schizophrenia patients [45]. When microglia activity in the cerebral cortex white matter of schizophrenia patients was assessed using (R)-[(11)C] PK11195 PET scanning (ABX, Radeberg, Germany), it was found to be increased, indicating the inflammatory response caused by microglia was tightly correlated with the onset of schizophrenia [46]. Notably, a recent study has shown that $Ca^{2+}$-related signaling pathways that play critical roles in the secretion of cytokines or inflammatory mediators produced by microglia may be main targets in the treatment of schizophrenia [46]. Similarly, the results of animal studies indicated that schizophrenia symptoms improved after treatment with minocycline, a type of tetracycline antibiotic which also acts as a microglial suppressor [47]. While it is possible that schizophrenia treatments or disease progression may cause inflammatory or abnormal immune responses in the brain, recent studies have consistently reported microglial dysfunction in schizophrenia patients. In a PET study, only patients examined within 5 years of the onset of schizophrenia showed activated microglia [46]. In a recent meta-analysis, relatively consistent changes in cytokine levels were

observed in schizophrenia patients regardless of the medication used; therefore, they are sometimes regarded as markers of schizophrenia [48].

According to an epidemiological study and supporting evidence from several other studies, a cause of schizophrenia may be infection with influenza virus during the second trimester of pregnancy [49]. Infection before birth enhances the sensitivity of the immune system to external stimuli, and overreaction of microglia becomes evident. A negative correlation was found between the onset of schizophrenia and rheumatic arthritis, while other autoimmune diseases showed a positive correlation, which indicated that abnormal immune system function could be a risk factor for schizophrenia [50]. Precisely how activated microglia cause abnormalities in neural connections and lead to psychotic symptoms of schizophrenia remains unclear. Nonetheless, a recent study with *DISC1* transgenic mice showed that activated microglia had an effect on GABA and dopamine signaling pathways [51] and that the cytokines and reactive oxygen species released by microglia had an effect on schizophrenia-like behavioral dysfunctions [52].

The reported increase in pro-inflammatory cytokines in schizophrenia suggests the possibility of novel drug development. Minocycline (tetracycline antibiotics) relies on a mechanism that controls microglia function, which ensures that it is an effective drug candidate for the treatment of schizophrenia. Minocycline can inhibit secretion of pro-inflammatory cytokines including IL-1, nitric oxide, and TNF-α and can block the nuclear translocation of nuclear factor kappa light-chain enhancer within activated B (NF-kappa B)-expressing cells [53, 54]. Several studies have reported a positive effect of minocycline in patients with schizophrenia or major depressive disorder [55]. Anti-inflammatory COX-2 inhibitors have also shown promising therapeutic effects in acute schizophrenia patients. In a study using a COX-2 inhibitor as an adjunct therapy in patients with schizophrenia, symptoms improved [56], although such effects were observed solely in acute schizophrenia patients with recent onset and not in chronic schizophrenia patients [57].

## 1.5 Neuron-Microglia Crosstalk in Neurocognitive Disorder

Microglia differ from neurons in that they maintain the ability to proliferate and regenerate even when mature [58]. The total number and density of microglia increase as animals or humans age, albeit with reduced immune functions (Table 1.1), such that the overall functional capacity remains similar; the increase in microglia numbers is thought to be the result of microglial proliferation and accumulation [59]. It is possible that the pathogenesis of AD is correlated with the morphological transformations of microglia caused by aging. In support of this hypothesis, accumulation of microglia around amyloid plaques has been reported in Alzheimer's disease (AD) [60]. Morphological changes in microglia in AD have also been reported. In general, the morphological transformations of microglia are characterized by an early contraction and subsequent hypertrophy to a slight degree [60]. The microglia in AD are typically short and thick with blunt protrusions, while

**Table 1.1** Neuron-microglia crosstalk in Alzheimer's disease

| Physiological functions of microglia | Abnormalities of microglia in Alzheimer's disease |
|---|---|
| Proliferation | • Increased total number and density of microglia<br>• Reduced immune functions of microglia<br>• Accumulation of microglia around plaques |
| Morphological transformation | • Dystrophy of microglia<br>• Loss of microglia protrusions and segmentation with an abnormally twisted cytoplasm |
| Motility and migration | • Increased concentrations of chemokines and their receptors that regulate microglial migration |
| Intercellular communication | • Microglia secrete various pro-inflammatory proteins<br>• Neural toxicity |
| Phagocytosis | • Impaired ability to degrade engulfed substances<br>• Not able to degrade myelin, Aβ, and cellular debris<br>• Reduced the rate of Aβ removal |

aged microglia generally lose their protrusions and become segmented with an abnormally twisted cytoplasm, thus displaying a pattern of dystrophy [61].

Microglial protrusions are considerably flexible to facilitate functional activity in the cerebral parenchyma. The mobility and migration of microglia are likely to be impaired as age increases [62]. In AD, newly emerging plaques are rapidly surrounded by microglia, and imaging studies found that β-amyloid (Aβ) was able to directly stimulate chemotaxis of microglia [63]: the concentration of chemokines and the corresponding receptors that regulate microglial migration increase, thus implying that Aβ deposition summons and activates the microglia. Notably, microglial migration toward the plaques is mediated by MCP-1, macrophage inflammatory protein (MIP)-1α, MIP-1β, IL-8, and macrophage-colony-stimulating factor (M-CSF) [64].

Microglia secrete various proteins with pro-inflammatory and anti-inflammatory functions. In AD patients and animal models, upregulation of several proteins in microglia has been reported and, in particular, that of human leukocyte antigen (HLA)-DR, cluster of differentiation (CD)11b/complement receptor type 3 (CR3), CD68, and toll-like receptors [65]. Secretion of these markers is generally correlated with antigen expression, lysosomal function, recognition of various pathogens, and complement proteins. These markers tend to increase in the brains of AD patients, which indicates a relative increase in neural toxicity as compared with neural protection [66]. Based on these findings, microglia in AD patients are considered to be constitutively active and stimulated.

Microglial cells are phagocytic, a process that includes recognition, engulfment, and degradation of a foreign substance in the CNS. The association between phagocytosis and AD has only been partially determined, although a study in rats showed the accumulation of various forms of vacuoles, vesicles, and lysosomal inclusions, which may be attributed to the impaired ability of microglia to degrade engulfed substances [67]. Aged microglia are not able to degrade myelin, Aβ, or cellular debris. Age and the stage of AD influence the phagocytotic ability of

microglia: in the early stages of AD, the removal of Aβ is efficient, but in later stages of AD, the rate of Aβ removal is reduced [68]. Based on this, future studies should focus on the improvement of microglia phagocytosis as an option for AD treatment.

Microglia exhibit simultaneous responses to various chemokines and cytokines related to Aβ. Several studies of AD patients have shown that microglia display pathological dysfunctions or reduced efficiency. A paradigm shift in AD treatment may therefore involve the development of drugs targeting the microglia-related neurotransmitters/chemokines and cytokines/inflammatory responses that mediate disease responses.

## 1.6 Conclusions

Neuron-microglia crosstalk has been shown to support close interactions among neurons or other types of neuroglia that play a critical role in maintaining normal behavior and cognitive function, rather than an assistive role in brain homeostasis or neurons. Postmortem examinations of brain tissue and animal studies recapitulating human mental disorders have also confirmed an association between psychiatric disorders and quantitative, structural, or functional abnormalities of neuron-microglia crosstalk. However, additional studies investigating the associations between microglial dysfunction and the symptoms of psychiatric disorders, as well studies to identify the specific underlying mechanisms of the changes in neurological function, are needed. The understanding of brain function and mental disorders requires consideration of neuron-microglia interactions, rather than solely focusing on the role of neurons. Future efforts to improve our understanding of the brain are thus predicted to initiate the search for the cause of psychiatric disorders and novel treatments.

## References

1. Funk GD, Rajani V, Alvares TS, Revill AL, Zhang Y, Chu NY, et al. Neuroglia and their roles in central respiratory control; an overview. Comp Biochem Physiol A Mol Integr Physiol. 2015;186:83–95.
2. Nishiyama A, Komitova M, Suzuki R, Zhu X. Polydendrocytes (NG2 cells): multifunctional cells with lineage plasticity. Nat Rev Neurosci. 2009;10(1):9–22.
3. Gundersen V, Storm-Mathisen J, Bergersen LH. Neuroglial transmission. Physiol Rev. 2015;95 (3):695–726.
4. Stevenson JA, Yoon MG. Mitosis of radial glial cells in the optic tectum of adult goldfish. J Neurosci. 1981;1(8):862–75.
5. Takebayashi M. Psychiatric disorders from a perspective of glia. Seishin Shinkeigaku Zasshi. 2013;115(12):1186–93.
6. Marques AH, Cizza G, Sternberg E. Brain-immune interactions and implications in psychiatric disorders. Braz J Psychiatry. 2007;29(Suppl 1):S27–32.
7. Prinz M, Jung S, Priller J. Microglia biology: one century of evolving concepts. Cell. 2019;179 (2):292–311.
8. Chen Z, Trapp BD. Microglia and neuroprotection. J Neurochem. 2016;136(Suppl 1):10–7.

9. Diaz-Aparicio I, Paris I, Sierra-Torre V, Plaza-Zabala A, Rodríguez-Iglesias N, Márquez-Ropero M, et al. Microglia actively remodel adult hippocampal neurogenesis through the phagocytosis secretome. J Neurosci. 2020;40(7):1453–82.
10. Lukacova N, Kisucka A, Kiss Bimbova K, Bacova M, Ileninova M, Kuruc T, et al. Glial-neuronal interactions in pathogenesis and treatment of spinal cord injury. Int J Mol Sci. 2021;22 (24):13577.
11. Garaschuk O, Verkhratsky A. Physiology of microglia. Methods Mol Biol. 2019;2034:27–40.
12. Zindler E, Zipp F. Neuronal injury in chronic CNS inflammation. Best Pract Res Clin Anaesthesiol. 2010;24(4):551–62.
13. Kirkham M, Berg DA, Simon A. Microglia activation during neuroregeneration in the adult vertebrate brain. Neurosci Lett. 2011;497(1):11–6.
14. Gómez-Palacio Schjetnan A, Escobar-Rodríguez ML. Memory coding and retention: brain-derived neurotrophic factor (BDNF) in synaptic plasticity. Rev Neurol. 2007;45(7):409–17.
15. Yuan C, Aierken A, Xie Z, Li N, Zhao J, Qing H. The age-related microglial transformation in Alzheimer's disease pathogenesis. Neurobiol Aging. 2020;92:82–91.
16. Wake H, Moorhouse AJ, Jinno S, Kohsaka S, Nabekura J. Resting microglia directly monitor the functional state of synapses in vivo and determine the fate of ischemic terminals. J Neurosci. 2009;29(13):3974–80.
17. Rajkowska G, Stockmeier CA. Astrocyte pathology in major depressive disorder: insights from human postmortem brain tissue. Curr Drug Targets. 2013;14(11):1225–36.
18. Williams MR, Sharma P, Fung KL, Pearce RK, Hirsch SR, Maier M. Axonal myelin increase in the callosal genu in depression but not schizophrenia. Psychol Med. 2015;45(10):2145–55.
19. Frick LR, Williams K, Pittenger C. Microglial dysregulation in psychiatric disease. Clin Dev Immunol. 2013;2013:608654.
20. Pan Y, Chen XY, Zhang QY, Kong LD. Microglial NLRP3 inflammasome activation mediates IL-1β-related inflammation in prefrontal cortex of depressive rats. Brain Behav Immun. 2014;41:90–100.
21. Tkachev D, Mimmack ML, Ryan MM, Wayland M, Freeman T, Jones PB, et al. Oligodendrocyte dysfunction in schizophrenia and bipolar disorder. Lancet. 2003;362(9386):798–805.
22. Watkins CC, Sawa A, Pomper MG. Glia and immune cell signaling in bipolar disorder: insights from neuropharmacology and molecular imaging to clinical application. Transl Psychiatry. 2014;4(1):e350.
23. Jia X, Gao Z, Hu H. Microglia in depression: current perspectives. Sci China Life Sci. 2021;64 (6):911–25.
24. Myint AM, Leonard BE, Steinbusch HW, Kim YK. Th1, Th2, and Th3 cytokine alterations in major depression. J Affect Disord. 2005;88(2):167–73.
25. Gogoleva VS, Drutskaya MS, Atretkhany KS. The role of microglia in the homeostasis of the central nervous system and Neuroinflammation. Mol Biol. 2019;53(5):790–8.
26. Jeon SW, Kim YK. Inflammation-induced depression: its pathophysiology and therapeutic implications. J Neuroimmunol. 2017;313:92–8.
27. Jeon SW, Kim YK. Neuroinflammation and cytokine abnormality in major depression: cause or consequence in that illness? World J Psychiatry. 2016;6(3):283–93.
28. Bay-Richter C, Linderholm KR, Lim CK, Samuelsson M, Träskman-Bendz L, Guillemin GJ, et al. A role for inflammatory metabolites as modulators of the glutamate N-methyl-D-aspartate receptor in depression and suicidality. Brain Behav Immun. 2015;43:110–7.
29. Kohler O, Krogh J, Mors O, Benros ME. Inflammation in depression and the potential for anti-inflammatory treatment. Curr Neuropharmacol. 2016;14(7):732–42.
30. Liu Y, Ho RC, Mak A. Interleukin (IL)-6, tumour necrosis factor alpha (TNF-α) and soluble interleukin-2 receptors (sIL-2R) are elevated in patients with major depressive disorder: a meta-analysis and meta-regression. J Affect Disord. 2012;139(3):230–9.
31. Janelidze S, Mattei D, Westrin Å, Träskman-Bendz L, Brundin L. Cytokine levels in the blood may distinguish suicide attempters from depressed patients. Brain Behav Immun. 2011;25(2): 335–9.

32. Ganança L, Oquendo MA, Tyrka AR, Cisneros-Trujillo S, Mann JJ, Sublette ME. The role of cytokines in the pathophysiology of suicidal behavior. Psychoneuroendocrinology. 2016;63: 296–310.
33. Hansen F, Battú CE, Dutra MF, Galland F, Lirio F, Broetto N, et al. Methylglyoxal and carboxyethyllysine reduce glutamate uptake and S100B secretion in the hippocampus independently of RAGE activation. Amino Acids. 2016;48(2):375–85.
34. Rothermundt M, Peters M, Prehn JH, Arolt V. S100B in brain damage and neurodegeneration. Microsc Res Tech. 2003;60(6):614–32.
35. Schroeter ML, Abdul-Khaliq H, Krebs M, Diefenbacher A, Blasig IE. Serum markers support disease-specific glial pathology in major depression. J Affect Disord. 2008;111(2-3):271–80.
36. Schroeter ML, Abdul-Khaliq H, Krebs M, Diefenbacher A, Blasig IE. Neuron-specific enolase is unaltered whereas S100B is elevated in serum of patients with schizophrenia—original research and meta-analysis. Psychiatry Res. 2009;167(1-2):66–72.
37. Kalia M, Costa ESJ. Biomarkers of psychiatric diseases: current status and future prospects. Metab Clin Exp. 2015;64(3 Suppl 1):S11–5.
38. Suchankova P, Klang J, Cavanna C, Holm G, Nilsson S, Jönsson EG, et al. Is the Gly82Ser polymorphism in the RAGE gene relevant to schizophrenia and the personality trait psychoticism? J Psychiatry Neurosci. 2012;37(2):122–8.
39. Kubicki M, McCarley RW, Shenton ME. Evidence for white matter abnormalities in schizophrenia. Curr Opin Psychiatry. 2005;18(2):121–34.
40. van den Heuvel MP, Sporns O, Collin G, Scheewe T, Mandl RC, Cahn W, et al. Abnormal rich club organization and functional brain dynamics in schizophrenia. JAMA Psychiat. 2013;70(8): 783–92.
41. Konrad A, Winterer G. Disturbed structural connectivity in schizophrenia primary factor in pathology or epiphenomenon? Schizophr Bull. 2008;34(1):72–92.
42. Pantazopoulos H, Woo TU, Lim MP, Lange N, Berretta S. Extracellular matrix-glial abnormalities in the amygdala and entorhinal cortex of subjects diagnosed with schizophrenia. Arch Gen Psychiatry. 2010;67(2):155–66.
43. Haybaeck J, Postruznik M, Miller CL, Dulay JR, Llenos IC, Weis S. Increased expression of retinoic acid-induced gene 1 in the dorsolateral prefrontal cortex in schizophrenia, bipolar disorder, and major depression. Neuropsychiatr Dis Treat. 2015;11:279–89.
44. Miyata S, Hattori T, Shimizu S, Ito A, Tohyama M. Disturbance of oligodendrocyte function plays a key role in the pathogenesis of schizophrenia and major depressive disorder. Biomed Res Int. 2015;2015:492367.
45. Balla P, Frecska E. Changes in the brain with schizophrenia: postmortem findings. Neuropsychopharmacol Hung. 2011;13(4):219–27.
46. van Berckel BN, Bossong MG, Boellaard R, Kloet R, Schuitemaker A, Caspers E, et al. Microglia activation in recent-onset schizophrenia: a quantitative (R)-[11C]PK11195 positron emission tomography study. Biol Psychiatry. 2008;64(9):820–2.
47. Mattei D, Djodari-Irani A, Hadar R, Pelz A, de Cossío LF, Goetz T, et al. Minocycline rescues decrease in neurogenesis, increase in microglia cytokines and deficits in sensorimotor gating in an animal model of schizophrenia. Brain Behav Immun. 2014;38:175–84.
48. Capuzzi E, Bartoli F, Crocamo C, Clerici M, Carrà G. Acute variations of cytokine levels after antipsychotic treatment in drug-naïve subjects with a first-episode psychosis: a meta-analysis. Neurosci Biobehav Rev. 2017;77:122–8.
49. Mednick SA, Machon RA, Huttunen MO, Bonett D. Adult schizophrenia following prenatal exposure to an influenza epidemic. Arch Gen Psychiatry. 1988;45(2):189–92.
50. Benros ME, Nielsen PR, Nordentoft M, Eaton WW, Dalton SO, Mortensen PB. Autoimmune diseases and severe infections as risk factors for schizophrenia: a 30-year population-based register study. Am J Psychiatry. 2011;168(12):1303–10.
51. Abazyan B, Nomura J, Kannan G, Ishizuka K, Tamashiro KL, Nucifora F, et al. Prenatal interaction of mutant DISC1 and immune activation produces adult psychopathology. Biol Psychiatry. 2010;68(12):1172–81.

52. Powell SB, Sejnowski TJ, Behrens MM. Behavioral and neurochemical consequences of cortical oxidative stress on parvalbumin-interneuron maturation in rodent models of schizophrenia. Neuropharmacology. 2012;62(3):1322–31.
53. Lai AY, Todd KG. Hypoxia-activated microglial mediators of neuronal survival are differentially regulated by tetracyclines. Glia. 2006;53(8):809–16.
54. Hovens JE, Onderwater TA. Minocycline for schizophrenia: a brief overview. Tijdschr Psychiatr. 2014;56(6):402–6.
55. Chaudhry IB, Hallak J, Husain N, Minhas F, Stirling J, Richardson P, et al. Minocycline benefits negative symptoms in early schizophrenia: a randomised double-blind placebo-controlled clinical trial in patients on standard treatment. J Psychopharmacol. 2012;26(9):1185–93.
56. Marini S, De Berardis D, Vellante F, Santacroce R, Orsolini L, Valchera A, et al. Celecoxib adjunctive treatment to antipsychotics in schizophrenia: a review of randomized clinical add-on trials. Mediators Inflamm. 2016;2016:3476240.
57. Rapaport MH, Delrahim KK, Bresee CJ, Maddux RE, Ahmadpour O, Dolnak D. Celecoxib augmentation of continuously ill patients with schizophrenia. Biol Psychiatry. 2005;57(12):1594–6.
58. Kaneko H, Nishiguchi KM, Nakamura M, Kachi S, Terasaki H. Characteristics of bone marrow-derived microglia in the normal and injured retina. Invest Ophthalmol Vis Sci. 2008;49(9):4162–8.
59. Streit WJ, Xue QS. The Brain's aging immune system. Aging Dis. 2010;1(3):254–61.
60. McGeer PL, Itagaki S, Tago H, McGeer EG. Reactive microglia in patients with senile dementia of the Alzheimer type are positive for the histocompatibility glycoprotein HLA-DR. Neurosci Lett. 1987;79(1-2):195–200.
61. Morales I, Farías G, Maccioni RB. Neuroimmunomodulation in the pathogenesis of Alzheimer's disease. Neuroimmunomodulation. 2010;17(3):202–4.
62. Damani MR, Zhao L, Fontainhas AM, Amaral J, Fariss RN, Wong WT. Age-related alterations in the dynamic behavior of microglia. Aging Cell. 2011;10(2):263–76.
63. Doens D, Fernández PL. Microglia receptors and their implications in the response to amyloid β for Alzheimer's disease pathogenesis. J Neuroinflammation. 2014;11:48.
64. McLarnon JG. Microglial chemotactic signaling factors in Alzheimer's disease. Am J Neurodegener Dis. 2012;1(3):199–204.
65. Landreth GE, Reed-Geaghan EG. Toll-like receptors in Alzheimer's disease. Curr Top Microbiol Immunol. 2009;336:137–53.
66. Mosher KI, Wyss-Coray T. Microglial dysfunction in brain aging and Alzheimer's disease. Biochem Pharmacol. 2014;88(4):594–604.
67. Tremblay M, Zettel ML, Ison JR, Allen PD, Majewska AK. Effects of aging and sensory loss on glial cells in mouse visual and auditory cortices. Glia. 2012;60(4):541–58.
68. Solito E, Sastre M. Microglia function in Alzheimer's disease. Front Pharmacol. 2012;3:14.

# 2 Microbiota–Gut–Brain Axis: Pathophysiological Mechanism in Neuropsychiatric Disorders

Cheolmin Shin and Yong-Ku Kim

**Abstract**

Gut microbiota influence human behavior. The immunological, metabolic, and endocrine systems are involved in bidirectional communication between the gut and the brain, which is regulated by microbes through the microbiota-derived neurochemicals and metabolites. Gut microbiota have certain effects on neurodevelopment and maturation of immunity. However, gut dysbiosis can lead to neuropsychiatric disorders. Animal research and clinical case-control studies have demonstrated that gut dysbiosis has an adverse effect on human behavior through a variety of mechanisms. Recent meta-analysis on clinical studies confirmed gut dysbiosis in several major neuropsychiatric disorders. Microbiota-targeted intervention has recently been in the spotlight and meta-analyses have confirmed its effectiveness. In this chapter, we summarize the evidence for the interactions between microbiota and brain–gut network, as well as the potential pathophysiological mechanisms involved.

**Keywords**

Microbiota–gut–brain axis · Microbiota · Gut-dysbiosis · Probiotics · Fecal microbiota transplantation

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## 2.1 Introduction

The gut has evolved to contain diverse microbial communities, including fungi, parasites, archaea, viruses, protozoa, and bacteria. The bacterial community is currently the most characterized. It is estimated that bacteria in humans are 1.3 times more numerous than human cells [1], and most are present in the gut. Gut microbiota are essential for maintaining health as they produce short-chain fatty acids (SCFAs), digest carbohydrates, synthesize vitamins, and metabolize toxins [2]. Of particular importance is their role in the maturation and development of the central nervous system (CNS), despite the gut microbiota being located away from the brain [3]. As a result, the gut microbiota has been demonstrated to affect mood, behavior, and cognition and is acknowledged to have an impact on the onset and progression of neuropsychiatric disorders [4].

Possible pathways to allow the brain and gut to interact with microbiota include neural (vagal and enteric), endocrine (hypothalamic–pituitary–adrenal [HPA] axis and enteroendocrine), metabolic (bacterial metabolites and host metabolism), and immunologic (innate and adaptive immunity) pathways (Fig. 2.1) [4, 5]. Each component involved in these pathways is not limited to one pathway but seems to be interrelated to the others. The exact mechanism of this communication is still under investigation; however, from the vast evidence gathered from animal, human

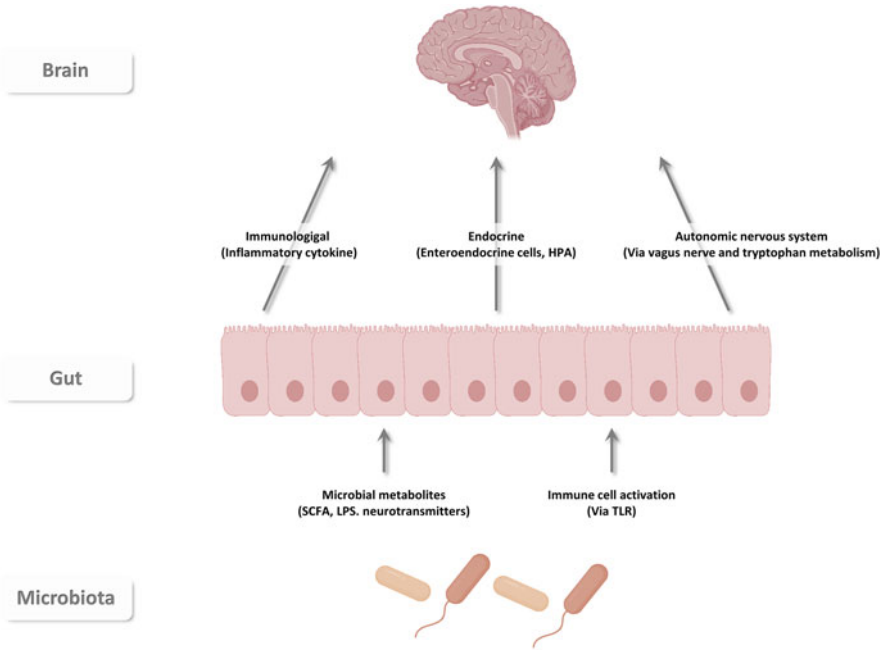


**Fig. 2.1** Communication pathways of the microbiota–gut–brain axis. *HPA* hypothalamic–pituitary–adrenal, *SCFA* short-chain fatty acids, *LPS* lipopolysaccharide, *TLR* toll-like receptor

cross-sectional, and human cohort studies, the importance of the gut microbiota in the interactions between the gut and the brain in both directions has been demonstrated.

In this chapter, we discussed how bacteria interacts with the brain–gut network and influences mental function, as well as how the microbiota–gut–brain axis affects neuropsychiatric disorders.

## 2.2 Microbiota–Gut–Brain Axis

It is widely acknowledged that resident microbiota can have a significant impact on host behavior [6, 7]. Bidirectional communication between gut and brain is an essential component of the synergy between the microbiota and host in accessing gut–brain signaling pathways to modify the host's brain and behavior [4]. Studies conducted to identify and investigate the microbiota–gut–brain axis have utilized a variety of interventions, including GF animals [8], antibiotic induction [9], prebiotic and probiotic supplementation [10, 11], pathogen infection in the gastrointestinal (GI) tract [12], and fecal microbiota transplantation (FMT) [13].

Gut dysbiosis has been linked to several negative outcomes in neuropsychiatric disorders [14–17]. Animal studies have shown that gut dysbiosis leads to a persistent low-grade pro-inflammatory state in the host by causing the intestinal barrier to become more permeable, making it easier for bacterial antigens to enter into the circulation [18]. This may lead to disease in those individuals prone to particular illnesses, whether in animals, experimental models, or otherwise healthy persons.

## 2.3 Potential Communication Pathways Between Gut Microbiota and the Brain

### 2.3.1 Immunological Pathway

Gut microbiota affect immunity by inducing the circulation of pro-inflammatory cells and cytokines through the interaction of microbial metabolites with intestinal host receptors and by directly interacting with host cells in the brain through systemic translocation of microbial metabolites. First, intestinal infection by microbiota can induce an inflammatory response directly in the CNS of host cells. Chronic low-grade inflammation further affects the immune system by releasing cytokines into the bloodstream. The gut microbiota contains pro-inflammatory substances such as lipopolysaccharides (LPSs) and peptidoglycans. LPS can be recognized by the toll-like receptor (TLR)-4, which is widely distributed in monocytes, macrophages, and microglia of the brain. Activation of the TLR-4-mediated inflammatory response by the gut microbiota has been reported in patients with inflammatory bowel syndrome and depression [19]. The indirect effects of gut microbiota on the innate immune system can directly affect brain function by altering pro-inflammatory and anti-inflammatory cytokines at the circulatory level

[20]. The commensal microbiota is known to influence the development of the host immune system [21] and affect the autoreactivity of peripheral immune cells to the host CNS [22]. Correlated and experimental data linking autoimmunity, GI activity, and neuropsychiatric disorders suggest a possible influence of the immune pathway on the pathogenesis of neuropsychiatric disorders.

### 2.3.2 Microbial Metabolites

The gut microbiota decomposes carbohydrates into various forms of SCFA, and the main components of SCFA produced in this way are acetate, propionate, and butyrate [23]. They are absorbed in the intestine and supplied to distant organs through blood vessels, where they are mainly used for production of the energy required for cell activity.

Butyric acid and propionic acid promote the expression of the genes for tyrosine hydroxylase, an enzyme that regulates the rate at which dopamine and noradrenaline are synthesized, and dopamine-β-hydroxylase, an enzyme that converts dopamine to noradrenaline [24]. GABA, serotonin, and dopamine levels were decreased in vivo by prolonged administration of propionic acid in germ-free rats [25]. SCFAs generated by microorganisms are consequently a part of a brain circuit that can influence physiology and behavior. Propionate generated by the intestinal microbiota boosted intestinal gluconeogenesis gene expression via a gut–brain neural circuit that included the fatty acid receptor FFAR3 [26].

SCFAs regulate the generation of inflammatory cytokines, chemokines, and lipid mediators by interacting with intestinal epithelial cells and immune cells such as neutrophils [27–29]. They also regulate gut barrier function and intestinal mucosal immunity [30]. By activating neutrophil receptors, acetate and propionate have been demonstrated to have an impact on the generation of circulating inflammatory cytokines and chemokines [31]. Before crossing the blood–brain barrier, SCFAs can reach the systemic circulation through the intestinal mucosa and impact innate immune cells, such as microglia and astrocytes in the brain [32]. G protein-coupled receptors (GPR), such as GPR41 and GPR43, frequently mediate interactions between SCFAs and innate immune cells [31, 33]. However, these receptors are not essential for SCFAs to reach the brain. Propionate, butyrate, and acetate seem to directly impact microglia through intracellular inhibition of histone deacetylases, which leads to increased transcription of certain genes involved in microglial function [32].

Numerous neurotransmitters and chemically related compounds can be produced by bacteria. Some gut bacterial strains have the ability to generate and locally release neurotransmitters such as γ-aminobutyric acid (GABA), serotonin, catecholamine, and histamine. Through enterochromaffin cells and enteric nerve receptors, these neurotransmitters originating from bacteria can transfer signals to the CNS.

In the human intestines, *Lactobacillus brevis* and *Bifidobacterium dentium* effectively manufacture GABA, a key inhibitory neurotransmitter in the central nervous system (CNS) whose malfunction is linked to sadness, anxiety, autism, and

schizophrenia [34]. In an animal investigation, Takanaga et al. hypothesized that GABA generated by gut bacteria penetrates the blood–brain barrier (BBB) and enters the CNS [35]. Mice exposed to *Lactobacillus rhamnosus* exhibited fewer depressive behaviors, and their hippocampuses contained more GABA [36, 37]. It is plausible that gut bacteria indirectly control GABA signaling via the vagus nerve given that such effects only appear when the vagus nerve is healthy.

The neuromodulators dopamine and noradrenaline play a key role in regulating vigilance, motivation, reward, learning, and memory processes. Given that SPF animals have significantly higher amounts of noradrenaline and dopamine in the cecum than germ-free mice do, it is likely that gut microbiota can provide catecholamine [38]. In some bacterial species, there is a gene for a transcript with a sequence similar to that of tyrosine hydroxylase, the rate-limiting enzyme in the synthesis of noradrenaline and dopamine [39]. Gut bacteria, such as *Enterococcus*, can produce dopamine [40]. Dopamine produced in the peripheral nervous system cannot cross the BBB; hence, the impact of catecholamines generated by microbes on the brain remains unproven. Tyrosine, the rate-limiting substrate for the synthesis of noradrenaline and dopamine, is found in lower concentrations in germ-free mice than in ex-germ-free mice, indicating that the gut microbiota may enhance dopamine levels in the brain [41]. A study comparing ex-germ-free animals to germ-free mice found that catecholamine levels were higher in the brains of the former group, but that gut microbiota restoration reduced those levels through regulating dopamine and noradrenaline reuptake in the brain [42].

Histamine, a neurotransmitter and immunomodulator, is involved in the control of circadian rhythm, cognition, waking, and neuroendocrine regulation. Histidine decarboxylase is expressed by *Lactobacillus reuteri*, which also produces histamine [43]. In addition to promoting histamine synthesis, *Lactobacillus reuteri* cultures also produce histidine decarboxylase. In addition, *Lactobacillus reuteri* suppresses the pro-inflammatory cytokine TNF-α via generating histamine in myeloid progenitor cells. Histamine has also been demonstrated to play an immunomodulatory role in the control of *Yersinia enterocolitica* infection in intestinal lymphoid organs [44].

### 2.3.3 Endocrine System

The gut is the largest endocrine organ, and epithelial enteroendocrine cells (EECs) are most important in brain–gut endocrine communication, although they account for less than 1% of intestinal endothelium [45]. The gut epithelium interacts directly with enteric neurons and epithelial cells via EECs in the presence of trophic and microbial stimuli [46, 47]. Certain subsets of the EEC also form neural circuits by synapsing directly with vagus neurons, enabling rapid communication with the brain by activating afferent synaptic transmission [48, 49]. There are various types of EECs that secrete 5-hydroxytryptamine (5-HT), regulatory peptides (e.g., glucagon-like peptides 1 [GLP-1], ghrelin, cholecystokinin, peptide YY), and bioactive molecules.

Exposure to bacterial metabolites, such as indole, activates EEC and induces 5-HT secretion, which stimulates the vagus sensory ganglion and activates cholinergic enteric neurons [50]. ECC directly detects gut microbiota and its metabolites by expressing a group of receptors including TRPA1 and TLR2 [49, 51]. Mucosal neurons were infected as a result of EECs delivering the rabies virus into the colon lumen [52]. This neuroepithelial circuit acted as a direct pathway for the nervous system to communicate with both food and gut bacteria.

GLP-1 receptor is currently being addressed in the context of mood regulation and neurodegenerative process [53, 54], in addition to its function in controlling eating behavior. Gut microbiota and microbial metabolites have been found to mediate the secretion and function of GLP-1, activating afferent nerve neurons in the colon in the process [55, 56].

The HPA axis is a major component of the neuroendocrine system that responds to stress [57], and the understanding of microbiota–brain communication through this axis is increasing. Stress affects the HPA axis by increasing the release of glucocorticoids, and its chronicity reduces hippocampal neurogenesis [58]. Chronic glucocorticoid exposure may lead to hippocampal fragility by diminishing neuronal differentiation and maturation via corticosterone [59]. It has been found that the hippocampal expression of glucocorticoid receptor pathway genes, which are responsible for intracellular cytokine receptor signaling, cellular growth, and neurotransmitter production, changes after the transplantation of gut microbiota from severely depressed patients into germ-free mice [60]. Microbiota modulation has been found to enhance social behavior and decrease corticosterone levels in mice subjected to high social stress by inhibiting HPA axis activation [61]. This effect was countered by antibiotic depletion of the gut microbiota and reversed by adrenalectomy, glucocorticoid receptor antagonists, and pharmacological suppression of corticosterone synthesis.

### 2.3.4 Autonomic Nervous System

The vagus nerve plays an important role in facilitating bidirectional communication between the CNS and the gut microbiota. A single synaptic link from the gut to the brain is made possible by enteroendocrine cells in the gut, which have been demonstrated to establish glutamatergic synapses with vagus nerve in the small and large intestine [62, 63]. Gut-innervated vagal afferents are important elements of the host reward circuitry, directly inducing the release of dopamine in the striatum [64]. All layers of the intestinal wall are covered by vagal afferent fibers; however, because they do not pass through the epithelium, they are unable to interact with the gut microbiota directly [65]. Furthermore, intestinal epithelial cells can produce peptides in response to bacterial metabolites, such as indoles. The peptides can be sensed indirectly by vagal afferent fibers through the diffusion of microbial metabolites [66]; the microbial synthesis of host molecules, such as gut serotonin [62]; and other mechanisms [50, 55]. The nucleus tractus solitarius relays vagus nerve stimulation to the brainstem and then to other regions of the brain [67], which

has implications for various clinical problems. In mice, a depression-like phenotype was observed according to the integrity of the vagus nerve, after the administration of *Lactobacillus reuteri* and LPS [68, 69]. *Lactobacillus reuteri* and *Lactobacillus intestinalis*, on the other hand, also provide advantageous neurological effects by reversing social behavioral abnormalities in ASD mice with an intact vagus nerve [70, 71]. The major impact of the vagotomy technique, which involves severing both afferent and efferent vagus nerve fibers and affects brain function, may contribute to the contradictory effects of *Lactobacillus* [72]. Additionally, compromising vagal integrity worsens inflammation [73], and this may account for the enhanced inflammatory activity of *Escherichia coli* and *Paenalcaligenes hominis* in mice with cognitive impairment post-vagotomy [74].

Tryptophan is a precursor of serotonin, which regulates the mood, cognition, and learning functions, and kynurenine, which is involved in the generation of neuroprotective and neurotoxic components [75]. A recent meta-analysis that confirmed the reduction of tryptophan and kynurenine in major depressive disorder (MDD), bipolar disorder, and schizophrenia underscored the importance of tryptophan metabolism in neuropsychiatric disorders [76]. Furthermore, its role has also been shown in autism spectrum disorders [77] and neurodegenerative diseases [78]. Gut microbiota has a significant impact on the availability of tryptophan, and research is ongoing to determine the link between alterations in microbiota functionality and various illnesses. According to the findings of prior studies, germ-free mice had greater serum levels of tryptophan and lower levels of serotonin than normally colonized mice, which may mean that tryptophan hydroxylase expression in the gut is lower in germ-free mice [79, 80]. *Bifidobacterium infantis* has been found to boost the levels of tryptophan and inflammatory indicators while decreasing the kynurenine-to-tryptophan ratio [81]. Changes in certain functional pathways involved in tryptophan production and metabolism have been discovered in recent investigations of intestinal metagenomes from patients with bipolar disorder with current major depressive episode [82].

The endocannabinoid system is an emerging pathway that is recognized as a modulator in the microbiota–gut–brain axis. The endocannabinoid system functions include modulating CNS responses to stressors through a signaling system that is composed of cannabinoid receptors, the mediator molecules such as the endogenous cannabinoid receptor ligands, and the enzymes involved in the production and degradation of these ligands [83]. The modulation of endocannabinoid system genes which are linked to gastrointestinal dysfunction and chronic stress has been linked to colonization studies in germ-free mouse models [84]. According to a cohort study that examined anhedonia and amotivation, people with more severe symptoms had higher amounts of palmitoylethanolamide, the endogenous cannabidiol, and less microbial diversity [85]. Fatty acid amide hydrolase, the primary catabolic enzyme of endocannabinoid agonists, is inhibited by palmitoylethanolamide. By controlling synaptic feedback to maintain excitatory and inhibitory balance in the CNS, endocannabinoid agonists protect mental health [86]. Chronic stress causes microbiota dysbiosis that alters lipid metabolism and endocannabinoid production,

which reduces adult neurogenesis in the hippocampus and endocannabinoid system signaling [87].

## 2.4 The Role of Gut–Microbiota–Brain Axis in Neuropsychiatric Illnesses

### 2.4.1 Autism Spectrum Disorder

Autism spectrum disorder (ASD) is a neurodevelopmental disorder characterized by deficiencies in social interaction and communication, as well as repetitive stereotyped behaviors. Chronic GI symptoms such as constipation and diarrhea are common in children with ASD [88]. Several studies have found that ASD severity is correlated with GI symptoms [89, 90], suggesting that the gut may significantly influence the onset and severity of ASD symptoms. In particular, the first evidence implicating the gut microbiota in ASD came from a small interventional trial in 11 children with regressive-onset autism, showing that oral vancomycin treatment improved both GI and ASD symptoms in these children [91].

Microbiological analysis has shown that the intestinal microflora of children with ASD is characterized by low diversity and is composed of an abnormal microbial community structure. Actinobacteria were abundant compared to the control. *Bacteroides, Parabacteroides, Clostridium, Faecalibacterium*, and *Phascolarctobacterium* were relatively abundant, whereas *Coprococcus* and *Bifidobacterium* accounted for a small proportion [92]. Some studies have reported no differences in the microbiota composition of children with ASD when compared with that in controls [93, 94].

### 2.4.2 Schizophrenia

Schizophrenia is a serious psychiatric illness that causes hallucinations, delusions, disorganized language and behavior, and negative symptoms. Epidemiologically, the risk of developing schizophrenia significantly increases following prenatal microbial infection [95]. Patients with schizophrenia have a higher incidence of intestinal barrier dysfunction, increased bacterial translocation, and more frequent GI diseases [96]. Patients with schizophrenia frequently have comorbidities associated with GI dysfunction, including inflammatory bowel diseases and celiac disease [97].

A cohort research found a nearly threefold elevated incidence of schizophrenia in individuals with soluble CD14, a sign of intestinal bacterial translocation [98]. Acute GI infection caused by *Toxoplasma gondii* causes an imbalance in the gut microbiota and a pro-inflammatory status with an elevated T cell response [99]. *Toxoplasma gondii* infection was reported as a risk factor for the development of early-onset schizophrenia in a cohort study [100].

A fecal microbiota analysis of a cohort of patients with first-episode psychotic disorders showed that increases in *Bifidobacterium* and *Lactobacillus* were associated with the severity of psychosis [101]. Patients with schizophrenia had less variety in their gut microbiota, and their disease severity was correlated with certain bacterial taxa including *Lachnospiraceae* and *Veillonellaceae* [102]. In this study, transplantation of microbiota from feces of schizophrenia patients lowered glutamate and increased GABA in the hippocampus and showed behaviors similar to animal models of schizophrenia involving glutamatergic hypofunction in germ-free mice.

In a clinical trial, *Lactobacillus rhamnosus* and *Bifidobacterium lactis* Bb12 were administered for 12 weeks in patients with schizophrenia, and they showed no change in the positive and negative syndrome scale (PANSS) score; however, they had decreased GI dysfunction [103]. In another clinical trial of patients with schizophrenia, probiotics containing *Lactobacillus acidophilus*, *Bifidobacterium bifidum, Lactobacillus reuteri,* and *Lactobacillus fermentum* significantly improved symptoms of schizophrenia, reduced inflammation, and increased plasma antioxidant capacity [104]. In another randomized controlled trial (RCT), PANSS and anxiety/depression scores were improved, and interferon-γ, IL-1R1, IL-10, and IL-22 levels were increased in 29 outpatients with schizophrenia who received *Bifidobacterium breve* A-1 for 4 weeks and decreased tumor necrosis factor-α levels [105].

### 2.4.3 Depression

Depression is a common mood disorder that causes severe symptoms that affect the way one feels, thinks, and processes daily activities, such as eating, sleeping, or working. Depressive symptoms are often accompanied by GI dysfunction such as inflammatory bowel disease and irritable bowel syndrome. Therefore, this may provide epidemiological evidence regarding the effects of gut microbiota on depression [106, 107].

HPA axis dysfunction is observed in patients with depression and in animal models of the disease [108]. Since animal studies have shown that modulation of the HPA axis is different in germ-free and wild-type mice, it is generally accepted that the gut microbiota may influence its effects [109].

The underlying mechanisms may include direct and indirect effects on the CNS of microbial metabolites such as SCFAs [110]. Changes in fecal SCFAs such as acetic acid, propionic acid, and pentanoic acid were observed in the animal depression model [111]. In humans with depressive symptoms, there was a positive relationship with acetate level and a negative relationship with both butyrate and propionate levels compared to healthy controls [112].

The "leaky gut" hypothesis for depression suggests that the translocation of bacteria by alterations in intestinal permeability leads to inflammation, resulting in symptoms similar to depression [113]. A positive correlation between a marker reflecting intestinal permeability and the Montgomery–Åsberg Depression Rating

Scale score was confirmed in patients with recent suicide attempts, patients with non-suicidal MDD, and healthy controls [114].

Changes in the gut microbiota have been described in individuals with depression. A meta-analysis found that the number of bacteria from the *Prevotellaceae* family and the genera *Coprococcus* and *Faecalibacterium* was lower in the gut of MDD patients than in the control group without depression [115]. A recent meta-analysis reported a depletion of certain anti-inflammatory butyrate-producing bacteria, such as *Faecalibacterium* and *Coprococcus* and an enrichment of pro-inflammatory bacteria, such as *Eggerthella* in patients with depression [116]. Implantation of gut microbiota obtained from depressed patients into mice and rats resulted in more depressed and anxious behavior than the control group that received microbes from healthy individuals, suggesting the possibility of a causal role of the depressive-gut microbiota in the development of depression [117, 118]. A meta-analysis of RCTs provided limited evidence of the benefit of adjunctive probiotics as compared to antidepressant monotherapy in the treatment of MDD [119].

### 2.4.4 Alzheimer's Disease

Alzheimer's disease (AD) is a neurodegenerative disorder characterized by progressive cognitive decline. Although the main question of how the microbiota–gut–brain axis contributes to the pathogenesis and/or progression of AD remains unclear, evidence suggests that the gut microbiota may be associated with the formation and processing of amyloid β (Aβ) and increased inflammatory reactions leading to neuronal death.

However, it is not yet known whether the gut microbiota has a direct effect on age-related cognitive decline. However, it may also play an important role in aging-related vulnerability. *Firmicutes* to *Bacteroidetes* ratio becomes more pronounced with advancing age, which may reflect inflammatory status [120]. It is likely that pathogenic bacteria increase with age at the expense of beneficial bacteria, which may be manifested by an increase in the relative abundance of *Proteobacteria* and decrease in *Bifidobacterium* species. This is also important because it can lead to chronic low-grade inflammation.

Systemic inflammation has the potential to lower the immunologically protective function of the brain and further promote the pathological progression of AD. An increased pro-inflammatory bacteria *Escherichia/Shigella* ratio and decreased levels of anti-inflammatory bacteria *Eubacterium rectale* are correlated with elevated levels of IL-1β, NLRP3, and CXCL2 in the plasma of patients with brain amyloidosis and cognitive impairment [121]. Gut microbiota also crosses the blood–brain barrier, producing pathogenic neurotoxins in the CNS through an inflammatory process that has a detrimental effect on the homeostatic function of neurons. The LPS was detected in the neocortex and hippocampus of patients with AD [122].

It has been proposed that Aβ accumulation and the microbiota–gut–brain axis are related. Increased *Firmicutes/Bacteroidetes* ratio and decreased trypsin were observed in transgenic mice to overproduce human amyloid precursor protein

(APP), suggesting that gut function is affected in AD-prone mice [123]. In APP/PS1 mice, which are APP overexpression mutant mice, the composition of gut microbiota changed with increasing age, which was also associated with increased Aβ levels and impairments in spatial learning and memory [124].

Several microbiota-targeted treatments have improved the progression of AD pathology and alleviated the symptoms of AD. APP/PS1 transgenic mice treated with probiotics such as *Bifidobacterium longum* and *Lactobacillus acidophilus* showed increased spatial memory and significantly decreased hippocampal plaques [125]. Long-term administration of probiotics containing *Bifidobacterium lactis*, *Lactobacillus casei*, *Bifidobacterium bifidum*, and *Lactobacillus acidophilus* resulted in significant improvements in fecal and brain microbial composition, cerebral nerve and synaptic damage, and immune response activation in mice with APP overproduction [126]. Probiotics also improved spatial cognitive impairment and significantly restored synaptic plasticity in rats intracerebroventricularly injected with Aβ [127]. One study examined the effects of microbiota-targeted treatment in humans, and patients with AD who received probiotics presented improved cognitive function and favorable changes in metabolic status, such as malondialdehyde and serum triglyceride [128]. Mice transplanted with fecal microbiota from wild-type mice to AD-like pathology with amyloid and neurofibrillary tangles ($ADLP^{APT}$) mice showed reduced formation, glial reactivity, and cognitive impairment of amyloid plaques and neurofibrillary tangles. Fecal microbiota transplant was thought to restore gut homeostasis in $ADLP^{APT}$ mice through aberrant expression of gut macrophage activity-related genes and reverse the increase in circulating blood inflammatory monocytes [129].

### 2.4.5 Parkinson's Disease

Parkinson's disease (PD) is a progressive neurodegenerative disease characterized by tremors, slow movements, rigidity, and distinctive gait. The destruction of nigral dopaminergic neurons and the development of Lewy bodies enriched in α-synuclein are cellular markers of PD [130]. GI dysregulation frequently precedes the onset of neurological symptoms of PD. Two decades ago, it was first proposed that PD starts in the gut and travels via the gut–brain axis to the brain [131].

Cell culture studies have shown that intestinal neurons release α-synuclein [132]. α-Synuclein is transported from the distal to the proximal vagus nerve in a time-dependent manner [133]. As a result, it is possible that, over time, any pathogenic activity that produces α-synuclein in the gut causes the illness to move to the brain. Indirect support for this theory comes from the accumulation of α-synuclein in rats that have bacteria which create extracellular bacterial amyloid proteins [134]. Interestingly, an epidemiological study found that individuals with duodenal ulcers who underwent vagotomy to eliminate the communication link between the brain and stomach axis had a decreased risk of PD [135].

The gut microbiota of patients with PD differed from that of healthy controls. The severity of movement-related symptoms such as postural instability and gait

disturbance was correlated with the prevalence of certain bacterial families [136]. Another study of gut microbiota from treatment-naive PD patients reported alteration in gut microbiota composition between PD patients and healthy controls [137]. *Bacteroidetes, Verrucomicrobia,* and *Proteobacteria* were more abundant in the feces of PD patients at the phylum level, and pro-inflammatory bacteria were more abundant at the genus level in them. Alterations in the gut microbiota caused by Parkinson's disease can result in significant functional differences that affect host metabolism and disease phenotype [138]. This study revealed that the predicted secretion potential of microbial metabolites, including increased methionine and cysteinylglycine, shows PD-associated metabolic patterns.

Microbiota-targeted interventions to treat PD symptoms are noteworthy. An animal study on a dietary supplement containing prebiotic fibers including galacto- and fructo-oligosaccharides (GOS and FOS, respectively) as well as other nutrients found benefit in motor, cognitive, and gastrointestinal (GI) symptoms in a mouse model of PD [139]. One clinical study published on the benefits of a fermented milk containing multiple strains of probiotics as well as prebiotics in treating constipation in PD [140].

## 2.5 Modulation of Gut Microbiota for the Treatment of Neuropsychiatric Disorders

Microbiota-targeted interventions have emerged as a promising avenue for the development of new therapeutic approaches due to the involvement of the gut microbiota in the process of mental illness and the possibility of modifying the microbiota through external factors [141]. Live microorganisms that have positive health effects on people or animals are referred to as probiotics [142]. Probiotics mostly consist of *Lactobacillus* and *Bifidobacterium* genera. Numerous positive findings have been published from animal studies that have examined the roles of probiotics in different neuropsychiatric conditions [143]. Recently published narrative and systematic reviews have investigated the benefits of probiotics on mental health outcomes [144], including particular diseases such as ASD [14], schizophrenia [145], and MDD [146]. Prebiotics are indigestible fibers that are selectively digested in the small intestine to support the development of *Lactobacillus* and *Bifidobacterium*, which are two healthy gut bacteria. GOS and FOS, inulins, and oligofructose are important prebiotics. The microbiota–gut–brain axis benefits from these prebiotics, which increase the amount of *Bifidobacterium* in the intestinal tract [147]. The composition of *Bacteroides* and *Bifidobacterium* appears to be normalized by the prebiotics in the healthy people [148]. Owing to the adjusted composition of the gut microbiota, GOS and FOS further increase the production of SCFAs [149]. However, evidence on the effectiveness of specific probiotics and prebiotics is currently limited.

Fecal microbiota transplantation (FMT) is the process of introducing healthy human feces into a patient diagnosed with probable gut dysbiosis to control the intestinal microbiota. The regeneration of normal bacterial flora by FMT can be

exceptional when the normal gut microbiota is eliminated by antibiotic treatment and *Clostridium difficile* enteritis develops [150]. Consequently, interest in FMT has increased. The first FMT trial in the neuropsychiatric field was recently published by Kang et al. Both GI and ASD symptoms were dramatically reduced in this 8-week, open-label clinical experiment to assess the effect of FMT on both symptoms. Eight weeks after treatment, the GI and ASD symptoms continued to improve [151].

## 2.6 Conclusions

The gut microbiota communicates with the brain, is involved in brain function, and inevitably affects the onset and course of brain disease. Although a clear association between microbiota and host physiology has been identified, a causal relationship has not yet been established. To clarify the processes by which the gut microbiota is responsible for neuropsychiatric disorders in humans, further human researches are required as many findings of brain–gut interactions have been gained from animal investigations. In particular, it emphasizes the importance of longitudinal large-scale human cohort studies, which will provide insight into the role of the gut microbiota, including genetic predisposition and environmental factors, such as prenatal exposure and life experience, in the investigation of the life cycle.

Although not yet perfect, microbiota-targeted therapies are promising approaches for the treatment of brain diseases. Most studies on the modulation of gut microbiota have been conducted on probiotics. According to a recent meta-analysis, probiotics for the treatment of neuropsychiatric disorders still lack specific evidence in terms of efficacy, and it is difficult to sufficiently explain their mode of action. FMT trials in humans are gradually being studied, and noteworthy results may be obtained in the future for specific subjects, such as those with treatment-resistant depression.

## References

1. Sender R, Fuchs S, Milo R. Revised estimates for the number of human and bacteria cells in the body. PLoS Biol. 2016;14(8):e1002533.
2. Valdes AM, Walter J, Segal E, Spector TD. Role of the gut microbiota in nutrition and health. BMJ. 2018;361:k2179.
3. Zheng D, Liwinski T, Elinav E. Interaction between microbiota and immunity in health and disease. Cell Res. 2020;30(6):492–506.
4. Cryan JF, Dinan TG. Mind-altering microorganisms: the impact of the gut microbiota on brain and behaviour. Nat Rev Neurosci. 2012;13(10):701–12.
5. Collins SM, Surette M, Bercik P. The interplay between the intestinal microbiota and the brain. Nat Rev Microbiol. 2012;10(11):735–42.
6. Clemente JC, Ursell LK, Parfrey LW, Knight R. The impact of the gut microbiota on human health: an integrative view. Cell. 2012;148(6):1258–70.
7. Karst SM. The influence of commensal bacteria on infection with enteric viruses. Nat Rev Microbiol. 2016;14(4):197–204.

8. Luczynski P, McVey Neufeld KA, Oriach CS, Clarke G, Dinan TG, Cryan JF. Growing up in a bubble: using germ-free animals to assess the influence of the gut microbiota on brain and behavior. Int J Neuropsychopharmacol. 2016;19(8):pyw020.
9. Desbonnet L, Clarke G, Traplin A, O'Sullivan O, Crispie F, Moloney RD, et al. Gut microbiota depletion from early adolescence in mice: implications for brain and behaviour. Brain Behav Immun. 2015;48:165–73.
10. Burokas A, Arboleya S, Moloney RD, Peterson VL, Murphy K, Clarke G, et al. Targeting the microbiota-gut-brain axis: prebiotics have anxiolytic and antidepressant-like effects and reverse the impact of chronic stress in mice. Biol Psychiatry. 2017;82(7):472–87.
11. Hemarajata P, Versalovic J. Effects of probiotics on gut microbiota: mechanisms of intestinal immunomodulation and neuromodulation. Therap Adv Gastroenterol. 2013;6(1):39–51.
12. Harris VC, Haak BW, Boele van Hensbroek M, Wiersinga WJ. The intestinal microbiome in infectious diseases: the clinical relevance of a rapidly emerging field. Open forum Infect Dis. 2017;4(3):ofx144.
13. Singh R, de Groot PF, Geerlings SE, Hodiamont CJ, Belzer C, Berge I, et al. Fecal microbiota transplantation against intestinal colonization by extended spectrum beta-lactamase producing Enterobacteriaceae: a proof of principle study. BMC Res Notes. 2018;11(1):190.
14. Ligezka AN, Sonmez AI, Corral-Frias MP, Golebiowski R, Lynch B, Croarkin PE, et al. A systematic review of microbiome changes and impact of probiotic supplementation in children and adolescents with neuropsychiatric disorders. Prog Neuropsychopharmacol Biol Psychiatry. 2021;108:110187.
15. Safadi JM, Quinton AMG, Lennox BR, Burnet PWJ, Minichino A. Gut dysbiosis in severe mental illness and chronic fatigue: a novel trans-diagnostic construct? A systematic review and meta-analysis. Mol Psychiatry. 2022;27(1):141–53.
16. Romano S, Savva GM, Bedarf JR, Charles IG, Hildebrand F, Narbad A. Meta-analysis of the Parkinson's disease gut microbiome suggests alterations linked to intestinal inflammation. Npj Parkinson's Dis. 2021;7(1):27.
17. Hung CC, Chang CC, Huang CW, Nouchi R, Cheng CH. Gut microbiota in patients with Alzheimer's disease spectrum: a systematic review and meta-analysis. Aging (Albany NY). 2022;14(1):477–96.
18. Kelly JR, Kennedy PJ, Cryan JF, Dinan TG, Clarke G, Hyland NP. Breaking down the barriers: the gut microbiome, intestinal permeability and stress-related psychiatric disorders. Front Cell Neurosci. 2015;9:392.
19. Daulatzai MA. Chronic functional bowel syndrome enhances gut-brain axis dysfunction, neuroinflammation, cognitive impairment, and vulnerability to dementia. Neurochem Res. 2014;39(4):624–44.
20. Cao P, Chen C, Liu A, Shan Q, Zhu X, Jia C, et al. Early-life inflammation promotes depressive symptoms in adolescence via microglial engulfment of dendritic spines. Neuron. 2021;109(16):2573–89 e9.
21. Kamada N, Seo SU, Chen GY, Nunez G. Role of the gut microbiota in immunity and inflammatory disease. Nat Rev Immunol. 2013;13(5):321–35.
22. Berer K, Krishnamoorthy G. Commensal gut flora and brain autoimmunity: a love or hate affair? Acta Neuropathol. 2012;123(5):639–51.
23. Lynch DB, Jeffery IB, Cusack S, O'Connor EM, O'Toole PW. Diet-microbiota-health interactions in older subjects: implications for healthy aging. Interdiscip Top Gerontol. 2015;40:141–54.
24. Nankova BB, Agarwal R, MacFabe DF, La Gamma EF. Enteric bacterial metabolites propionic and butyric acid modulate gene expression, including CREB-dependent catecholaminergic neurotransmission, in PC12 cells—possible relevance to autism spectrum disorders. PLoS One. 2014;9(8):e103740.
25. El-Ansary AK, Ben Bacha A, Kotb M. Etiology of autistic features: the persisting neurotoxic effects of propionic acid. J Neuroinflammation. 2012;9:74.

26. De Vadder F, Kovatcheva-Datchary P, Goncalves D, Vinera J, Zitoun C, Duchampt A, et al. Microbiota-generated metabolites promote metabolic benefits via gut-brain neural circuits. Cell. 2014;156(1–2):84–96.
27. Correa-Oliveira R, Fachi JL, Vieira A, Sato FT, Vinolo MA. Regulation of immune cell function by short-chain fatty acids. Clin Transl Immunology. 2016;5(4):e73.
28. Parada Venegas D, De la Fuente MK, Landskron G, Gonzalez MJ, Quera R, Dijkstra G, et al. Short chain fatty acids (SCFAs)-mediated gut epithelial and immune regulation and its relevance for inflammatory bowel diseases. Front Immunol. 2019;10:277.
29. Kim CH. Control of lymphocyte functions by gut microbiota-derived short-chain fatty acids. Cell Mol Immunol. 2021;18(5):1161–71.
30. Rodrigues HG, Takeo Sato F, Curi R, Vinolo MAR. Fatty acids as modulators of neutrophil recruitment, function and survival. Eur J Pharmacol. 2016;785:50–8.
31. Kim MH, Kang SG, Park JH, Yanagisawa M, Kim CH. Short-chain fatty acids activate GPR41 and GPR43 on intestinal epithelial cells to promote inflammatory responses in mice. Gastroenterology. 2013;145(2):396–406 e1–10.
32. Erny D, Hrabe de Angelis AL, Jaitin D, Wieghofer P, Staszewski O, David E, et al. Host microbiota constantly control maturation and function of microglia in the CNS. Nat Neurosci. 2015;18(7):965–77.
33. Yang G, Chen S, Deng B, Tan C, Deng J, Zhu G, et al. Implication of G protein-coupled receptor 43 in intestinal inflammation: a mini-review. Front Immunol. 2018;9:1434.
34. Barrett E, Ross RP, O'Toole PW, Fitzgerald GF, Stanton C. γ-Aminobutyric acid production by culturable bacteria from the human intestine. J Appl Microbiol. 2012;113(2):411–7.
35. Takanaga H, Ohtsuki S, Hosoya K, Terasaki T. GAT2/BGT-1 as a system responsible for the transport of gamma-aminobutyric acid at the mouse blood-brain barrier. J Cereb Blood Flow Metab. 2001;21(10):1232–9.
36. Bravo JA, Forsythe P, Chew MV, Escaravage E, Savignac HM, Dinan TG, et al. Ingestion of lactobacillus strain regulates emotional behavior and central GABA receptor expression in a mouse via the vagus nerve. Proc Natl Acad Sci U S A. 2011;108(38):16050–5.
37. Janik R, Thomason LAM, Stanisz AM, Forsythe P, Bienenstock J, Stanisz GJ. Magnetic resonance spectroscopy reveals oral lactobacillus promotion of increases in brain GABA, N-acetyl aspartate and glutamate. Neuroimage. 2016;125:988–95.
38. Asano Y, Hiramoto T, Nishino R, Aiba Y, Kimura T, Yoshihara K, et al. Critical role of gut microbiota in the production of biologically active, free catecholamines in the gut lumen of mice. Am J Physiol Gastrointest Liver Physiol. 2012;303(11):G1288–95.
39. Hernández-Romero D, Sanchez-Amat A, Solano F. A tyrosinase with an abnormally high tyrosine hydroxylase/dopa oxidase ratio. FEBS J. 2006;273(2):257–70.
40. Divyashri G, Krishna G, Muralidhara PSG. Probiotic attributes, antioxidant, anti-inflammatory and neuromodulatory effects of enterococcus faecium CFR 3003: in vitro and in vivo evidence. J Med Microbiol. 2015;64(12):1527–40.
41. Matsumoto M, Kibe R, Ooga T, Aiba Y, Sawaki E, Koga Y, et al. Cerebral low-molecular metabolites influenced by intestinal microbiota: a pilot study. Front Syst Neurosci. 2013;7:9.
42. Nishino R, Mikami K, Takahashi H, Tomonaga S, Furuse M, Hiramoto T, et al. Commensal microbiota modulate murine behaviors in a strictly contamination-free environment confirmed by culture-based methods. Neurogastroenterol Motil. 2013;25(6):521–8.
43. Thomas CM, Hong T, van Pijkeren JP, Hemarajata P, Trinh DV, Hu W, et al. Histamine derived from probiotic lactobacillus reuteri suppresses TNF via modulation of PKA and ERK signaling. PLoS One. 2012;7(2):e31951.
44. Handley SA, Dube PH, Miller VL. Histamine signaling through the H(2) receptor in the Peyer's patch is important for controlling yersinia enterocolitica infection. Proc Natl Acad Sci U S A. 2006;103(24):9268–73.
45. Latorre R, Sternini C, De Giorgio R, Greenwood-Van MB. Enteroendocrine cells: a review of their role in brain-gut communication. Neurogastroenterol Motil. 2016;28(5):620–30.

46. Bertrand PP, Bertrand RL. Serotonin release and uptake in the gastrointestinal tract. Auton Neurosci. 2010;153(1–2):47–57.
47. Bohorquez DV, Shahid RA, Erdmann A, Kreger AM, Wang Y, Calakos N, et al. Neuroepithelial circuit formed by innervation of sensory enteroendocrine cells. J Clin Invest. 2015;125(2):782–6.
48. Williams EK, Chang RB, Strochlic DE, Umans BD, Lowell BB, Liberles SD. Sensory neurons that detect stretch and nutrients in the digestive system. Cell. 2016;166(1):209–21.
49. Bellono NW, Bayrer JR, Leitch DB, Castro J, Zhang C, O'Donnell TA, et al. Enterochromaffin cells are gut chemosensors that couple to sensory neural pathways. Cell. 2017;170(1): 185–98 e16.
50. Ye L, Bae M, Cassilly CD, Jabba SV, Thorpe DW, Martin AM, et al. Enteroendocrine cells sense bacterial tryptophan catabolites to activate enteric and vagal neuronal pathways. Cell Host Microbe. 2021;29(2):179–96 e9.
51. Wang H, Kwon YH, Dewan V, Vahedi F, Syed S, Fontes ME, et al. TLR2 plays a pivotal role in mediating mucosal serotonin production in the gut. J Immunol. 2019;202(10):3041–52.
52. Bohórquez DV, Shahid RA, Erdmann A, Kreger AM, Wang Y, Calakos N, et al. Neuroepithelial circuit formed by innervation of sensory enteroendocrine cells. J Clin Invest. 2015;125(2):782–6.
53. Kim YK, Kim OY, Song J. Alleviation of depression by glucagon-like peptide 1 through the regulation of neuroinflammation, neurotransmitters, neurogenesis, and synaptic function. Front Pharmacol. 2020;11:1270.
54. Corbett A, Pickett J, Burns A, Corcoran J, Dunnett SB, Edison P, et al. Drug repositioning for Alzheimer's disease. Nat Rev Drug Discov. 2012;11(11):833–46.
55. Buckley MM, O'Brien R, Brosnan E, Ross RP, Stanton C, Buckley JM, et al. Glucagon-like Peptide-1 secreting L-cells coupled to sensory nerves translate microbial signals to the host rat nervous system. Front Cell Neurosci. 2020;14:95.
56. Martchenko SE, Martchenko A, Cox BJ, Naismith K, Waller A, Gurges P, et al. Circadian GLP-1 secretion in mice is dependent on the intestinal microbiome for maintenance of diurnal metabolic homeostasis. Diabetes. 2020;69(12):2589–602.
57. Russell G, Lightman S. The human stress response. Nat Rev Endocrinol. 2019;15(9):525–34.
58. Gould E, Tanapat P. Stress and hippocampal neurogenesis. Biol Psychiatry. 1999;46(11): 1472–9.
59. Levone BR, Codagnone MG, Moloney GM, Nolan YM, Cryan JF, Olivia OF. Adult-born neurons from the dorsal, intermediate, and ventral regions of the longitudinal axis of the hippocampus exhibit differential sensitivity to glucocorticoids. Mol Psychiatry. 2021;26(7): 3240–52.
60. Luo Y, Zeng B, Zeng L, Du X, Li B, Huo R, et al. Gut microbiota regulates mouse behaviors through glucocorticoid receptor pathway genes in the hippocampus. Transl Psychiatry. 2018;8 (1):187.
61. Wu WL, Adame MD, Liou CW, Barlow JT, Lai TT, Sharon G, et al. Microbiota regulate social behaviour via stress response neurons in the brain. Nature. 2021;595(7867):409–14.
62. Fulling C, Dinan TG, Cryan JF. Gut microbe to brain signaling: what happens in vagus. Neuron. 2019;101(6):998–1002.
63. Kaelberer MM, Buchanan KL, Klein ME, Barth BB, Montoya MM, Shen X, et al. A gut-brain neural circuit for nutrient sensory transduction. Science. 2018;361(6408):eaat5236.
64. Han W, Tellez LA, Perkins MH, Perez IO, Qu T, Ferreira J, et al. A neural circuit for gut-induced reward. Cell. 2018;175(3):665–78 e23.
65. Wang FB, Powley TL. Vagal innervation of intestines: afferent pathways mapped with new en bloc horseradish peroxidase adaptation. Cell Tissue Res. 2007;329(2):221–30.
66. Raybould HE. Gut chemosensing: interactions between gut endocrine cells and visceral afferents. Auton Neurosci. 2010;153(1–2):41–6.

67. Goehler LE, Gaykema RP, Opitz N, Reddaway R, Badr N, Lyte M. Activation in vagal afferents and central autonomic pathways: early responses to intestinal infection with campylobacter jejuni. Brain Behav Immun. 2005;19(4):334–44.
68. Wang S, Ishima T, Zhang J, Qu Y, Chang L, Pu Y, et al. Ingestion of lactobacillus intestinalis and lactobacillus reuteri causes depression- and anhedonia-like phenotypes in antibiotic-treated mice via the vagus nerve. J Neuroinflammation. 2020;17(1):241.
69. Zhang J, Ma L, Chang L, Pu Y, Qu Y, Hashimoto K. A key role of the subdiaphragmatic vagus nerve in the depression-like phenotype and abnormal composition of gut microbiota in mice after lipopolysaccharide administration. Transl Psychiatry. 2020;10(1):186.
70. Sgritta M, Dooling SW, Buffington SA, Momin EN, Francis MB, Britton RA, et al. Mechanisms underlying microbial-mediated changes in social behavior in mouse models of autism spectrum disorder. Neuron. 2019;101(2):246–59 e6.
71. Buffington SA, Dooling SW, Sgritta M, Noecker C, Murillo OD, Felice DF, et al. Dissecting the contribution of host genetics and the microbiome in complex behaviors. Cell. 2021;184(7):1740–56 e16.
72. Liu Y, Forsythe P. Vagotomy and insights into the microbiota-gut-brain axis. Neurosci Res. 2021;168:20–7.
73. Liu Y, Sanderson D, Mian MF, McVey Neufeld KA, Forsythe P. Loss of vagal integrity disrupts immune components of the microbiota-gut-brain axis and inhibits the effect of lactobacillus rhamnosus on behavior and the corticosterone stress response. Neuropharmacology. 2021;195:108682.
74. Lee KE, Kim JK, Han SK, Lee DY, Lee HJ, Yim SV, et al. The extracellular vesicle of gut microbial Paenalcaligenes hominis is a risk factor for vagus nerve-mediated cognitive impairment. Microbiome. 2020;8(1):107.
75. O'Mahony SM, Clarke G, Borre YE, Dinan TG, Cryan JF. Serotonin, tryptophan metabolism and the brain-gut-microbiome axis. Behav Brain Res. 2015;277:32–48.
76. Marx W, McGuinness AJ, Rocks T, Ruusunen A, Cleminson J, Walker AJ, et al. The kynurenine pathway in major depressive disorder, bipolar disorder, and schizophrenia: a meta-analysis of 101 studies. Mol Psychiatry. 2021;26(8):4158–78.
77. Bryn V, Verkerk R, Skjeldal OH, Saugstad OD, Ormstad H. Kynurenine pathway in autism spectrum disorders in children. Neuropsychobiology. 2017;76(2):82–8.
78. Maddison DC, Giorgini F. The kynurenine pathway and neurodegenerative disease. Semin Cell Dev Biol. 2015;40:134–41.
79. Yano JM, Yu K, Donaldson GP, Shastri GG, Ann P, Ma L, et al. Indigenous bacteria from the gut microbiota regulate host serotonin biosynthesis. Cell. 2015;161(2):264–76.
80. Wikoff WR, Anfora AT, Liu J, Schultz PG, Lesley SA, Peters EC, et al. Metabolomics analysis reveals large effects of gut microflora on mammalian blood metabolites. Proc Natl Acad Sci U S A. 2009;106(10):3698–703.
81. Desbonnet L, Garrett L, Clarke G, Bienenstock J, Dinan TG. The probiotic Bifidobacteria infantis: an assessment of potential antidepressant properties in the rat. J Psychiatr Res. 2008;43(2):164–74.
82. Lai WT, Zhao J, Xu SX, Deng WF, Xu D, Wang MB, et al. Shotgun metagenomics reveals both taxonomic and tryptophan pathway differences of gut microbiota in bipolar disorder with current major depressive episode patients. J Affect Disord. 2021;278:311–9.
83. Cani PD, Plovier H, Van Hul M, Geurts L, Delzenne NM, Druart C, et al. Endocannabinoids—at the crossroads between the gut microbiota and host metabolism. Nat Rev Endocrinol. 2016;12(3):133–43.
84. Sharkey KA, Wiley JW. The role of the endocannabinoid system in the brain-gut axis. Gastroenterology. 2016;151(2):252–66.
85. Minichino A, Jackson MA, Francesconi M, Steves CJ, Menni C, Burnet PWJ, et al. Endocannabinoid system mediates the association between gut-microbial diversity and anhedonia/amotivation in a general population cohort. Mol Psychiatry. 2021;26(11):6269–76.

86. Minichino A, Senior M, Brondino N, Zhang SH, Godwlewska BR, Burnet PWJ, et al. Measuring disturbance of the endocannabinoid system in psychosis: a systematic review and meta-analysis. JAMA Psychiat. 2019;76(9):914–23.
87. Chevalier G, Siopi E, Guenin-Macé L, Pascal M, Laval T, Rifflet A, et al. Effect of gut microbiota on depressive-like behaviors in mice is mediated by the endocannabinoid system. Nat Commun. 2020;11(1):6363.
88. McElhanon BO, McCracken C, Karpen S, Sharp WG. Gastrointestinal symptoms in autism spectrum disorder: a meta-analysis. Pediatrics. 2014;133(5):872–83.
89. Adams JB, Johansen LJ, Powell LD, Quig D, Rubin RA. Gastrointestinal flora and gastrointestinal status in children with autism—comparisons to typical children and correlation with autism severity. BMC Gastroenterol. 2011;11:22.
90. Chaidez V, Hansen RL, Hertz-Picciotto I. Gastrointestinal problems in children with autism, developmental delays or typical development. J Autism Dev Disord. 2014;44(5):1117–27.
91. Sandler RH, Finegold SM, Bolte ER, Buchanan CP, Maxwell AP, Vaisanen ML, et al. Short-term benefit from oral vancomycin treatment of regressive-onset autism. J Child Neurol. 2000;15(7):429–35.
92. Iglesias-Vazquez L, Van Ginkel RG, Arija V, Canals J. Composition of gut microbiota in children with autism spectrum disorder: a systematic review and meta-analysis. Nutrients. 2020;12(3):792.
93. Gondalia SV, Palombo EA, Knowles SR, Cox SB, Meyer D, Austin DW. Molecular characterisation of gastrointestinal microbiota of children with autism (with and without gastrointestinal dysfunction) and their neurotypical siblings. Autism Res. 2012;5(6):419–27.
94. Son JS, Zheng LJ, Rowehl LM, Tian X, Zhang Y, Zhu W, et al. Comparison of fecal microbiota in children with autism spectrum disorders and neurotypical siblings in the simons simplex collection. PLoS One. 2015;10(10):e0137725.
95. Brown AS, Derkits EJ. Prenatal infection and schizophrenia: a review of epidemiologic and translational studies. Am J Psychiatry. 2010;167(3):261–80.
96. Golofast B, Vales K. The connection between microbiome and schizophrenia. Neurosci Biobehav Rev. 2020;108:712–31.
97. West J, Logan RF, Hubbard RB, Card TR. Risk of schizophrenia in people with coeliac disease, ulcerative colitis and Crohn's disease: a general population-based study. Aliment Pharmacol Ther. 2006;23(1):71–4.
98. Severance EG, Gressitt KL, Stallings CR, Origoni AE, Khushalani S, Leweke FM, et al. Discordant patterns of bacterial translocation markers and implications for innate immune imbalances in schizophrenia. Schizophr Res. 2013;148(1–3):130–7.
99. Hand TW, Dos Santos LM, Bouladoux N, Molloy MJ, Pagan AJ, Pepper M, et al. Acute gastrointestinal infection induces long-lived microbiota-specific T cell responses. Science. 2012;337(6101):1553–6.
100. Mortensen PB, Nørgaard-Pedersen B, Waltoft BL, Sørensen TL, Hougaard D, Torrey EF, et al. Toxoplasma gondii as a risk factor for early-onset schizophrenia: analysis of filter paper blood samples obtained at birth. Biol Psychiatry. 2007;61(5):688–93.
101. Schwarz E, Maukonen J, Hyytiainen T, Kieseppa T, Oresic M, Sabunciyan S, et al. Analysis of microbiota in first episode psychosis identifies preliminary associations with symptom severity and treatment response. Schizophr Res. 2018;192:398–403.
102. Zheng P, Zeng B, Liu M, Chen J, Pan J, Han Y, et al. The gut microbiome from patients with schizophrenia modulates the glutamate-glutamine-GABA cycle and schizophrenia-relevant behaviors in mice. Sci Adv. 2019;5(2):eaau8317.
103. Dickerson FB, Stallings C, Origoni A, Katsafanas E, Savage CL, Schweinfurth LA, et al. Effect of probiotic supplementation on schizophrenia symptoms and association with gastrointestinal functioning: a randomized, placebo-controlled trial. Prim Care Companion CNS Disord. 2014;16(1):PCC.13m01579.

104. Ghaderi A, Banafshe HR, Mirhosseini N, Moradi M, Karimi MA, Mehrzad F, et al. Clinical and metabolic response to vitamin D plus probiotic in schizophrenia patients. BMC Psychiatry. 2019;19(1):77.
105. Okubo R, Koga M, Katsumata N, Odamaki T, Matsuyama S, Oka M, et al. Effect of bifidobacterium breve A-1 on anxiety and depressive symptoms in schizophrenia: a proof-of-concept study. J Affect Disord. 2019;245:377–85.
106. Bhandari S, Larson ME, Kumar N, Stein D. Association of inflammatory bowel disease (IBD) with depressive symptoms in the United States population and independent predictors of depressive symptoms in an IBD population: a NHANES study. Gut Liver. 2017;11(4):512–9.
107. Fond G, Loundou A, Hamdani N, Boukouaci W, Dargel A, Oliveira J, et al. Anxiety and depression comorbidities in irritable bowel syndrome (IBS): a systematic review and meta-analysis. Eur Arch Psychiatry Clin Neurosci. 2014;264(8):651–60.
108. Willner P, Scheel-Kruger J, Belzung C. The neurobiology of depression and antidepressant action. Neurosci Biobehav Rev. 2013;37(10 Pt 1):2331–71.
109. Sudo N, Chida Y, Aiba Y, Sonoda J, Oyama N, Yu XN, et al. Postnatal microbial colonization programs the hypothalamic-pituitary-adrenal system for stress response in mice. J Physiol. 2004;558(Pt 1):263–75.
110. Caspani G, Kennedy S, Foster JA, Swann J. Gut microbial metabolites in depression: understanding the biochemical mechanisms. Microb Cell. 2019;6(10):454–81.
111. Wu M, Tian T, Mao Q, Zou T, Zhou C-j, Xie J, et al. Associations between disordered gut microbiota and changes of neurotransmitters and short-chain fatty acids in depressed mice. Transl Psychiatry. 2020;10(1):350.
112. Müller B, Rasmusson AJ, Just D, Jayarathna S, Moazzami A, Novicic ZK, et al. Fecal short-chain fatty acid ratios as related to gastrointestinal and depressive symptoms in young adults. Psychosom Med. 2021;83(7):693–9.
113. Maes M, Kubera M, Leunis JC. The gut-brain barrier in major depression: intestinal mucosal dysfunction with an increased translocation of LPS from gram negative enterobacteria (leaky gut) plays a role in the inflammatory pathophysiology of depression. Neuro Endocrinol Lett. 2008;29(1):117–24.
114. Ohlsson L, Gustafsson A, Lavant E, Suneson K, Brundin L, Westrin A, et al. Leaky gut biomarkers in depression and suicidal behavior. Acta Psychiatr Scand. 2019;139(2):185–93.
115. Sanada K, Nakajima S, Kurokawa S, Barcelo-Soler A, Ikuse D, Hirata A, et al. Gut microbiota and major depressive disorder: a systematic review and meta-analysis. J Affect Disord. 2020;266:1–13.
116. Nikolova VL, Hall MRB, Hall LJ, Cleare AJ, Stone JM, Young AH. Perturbations in gut microbiota composition in psychiatric disorders: a review and meta-analysis. JAMA Psychiat. 2021;78(12):1343–54.
117. Zheng P, Zeng B, Zhou C, Liu M, Fang Z, Xu X, et al. Gut microbiome remodeling induces depressive-like behaviors through a pathway mediated by the host's metabolism. Mol Psychiatry. 2016;21(6):786–96.
118. Kelly JR, Borre Y, Ciaran OB, Patterson E, El Aidy S, Deane J, et al. Transferring the blues: depression-associated gut microbiota induces neurobehavioural changes in the rat. J Psychiatr Res. 2016;82:109–18.
119. Nikolova V, Zaidi SY, Young AH, Cleare AJ, Stone JM. Gut feeling: randomized controlled trials of probiotics for the treatment of clinical depression: systematic review and meta-analysis. Ther Adv Psychopharmacol. 2019;9:2045125319859963.
120. Biagi E, Nylund L, Candela M, Ostan R, Bucci L, Pini E, et al. Through ageing, and beyond: gut microbiota and inflammatory status in seniors and centenarians. PLoS One. 2010;5(5):e10667.
121. Cattaneo A, Cattane N, Galluzzi S, Provasi S, Lopizzo N, Festari C, et al. Association of brain amyloidosis with pro-inflammatory gut bacterial taxa and peripheral inflammation markers in cognitively impaired elderly. Neurobiol Aging. 2017;49:60–8.

122. Zhao Y, Jaber V, Lukiw WJ. Secretory products of the human GI tract microbiome and their potential impact on Alzheimer's disease (AD): detection of lipopolysaccharide (LPS) in AD hippocampus. Front Cell Infect Microbiol. 2017;7:318.
123. Brandscheid C, Schuck F, Reinhardt S, Schafer KH, Pietrzik CU, Grimm M, et al. Altered gut microbiome composition and tryptic activity of the 5xFAD Alzheimer's mouse model. J Alzheimers Dis. 2017;56(2):775–88.
124. Shen L, Liu L, Ji HF. Alzheimer's disease histological and behavioral manifestations in transgenic mice correlate with specific gut microbiome state. J Alzheimers Dis. 2017;56(1): 385–90.
125. Abraham D, Feher J, Scuderi GL, Szabo D, Dobolyi A, Cservenak M, et al. Exercise and probiotics attenuate the development of Alzheimer's disease in transgenic mice: role of microbiome. Exp Gerontol. 2019;115:122–31.
126. Yang X, Yu D, Xue L, Li H, Du J. Probiotics modulate the microbiota-gut-brain axis and improve memory deficits in aged SAMP8 mice. Acta Pharm Sin B. 2020;10(3):475–87.
127. Rezaei Asl Z, Sepehri G, Salami M. Probiotic treatment improves the impaired spatial cognitive performance and restores synaptic plasticity in an animal model of Alzheimer's disease. Behav Brain Res. 2019;376:112183.
128. Akbari E, Asemi Z, Daneshvar Kakhaki R, Bahmani F, Kouchaki E, Tamtaji OR, et al. Effect of probiotic supplementation on cognitive function and metabolic status in Alzheimer's disease: a randomized, double-blind and controlled trial. Front Aging Neurosci. 2016;8:256.
129. Kim MS, Kim Y, Choi H, Kim W, Park S, Lee D, et al. Transfer of a healthy microbiota reduces amyloid and tau pathology in an Alzheimer's disease animal model. Gut. 2020;69(2): 283–94.
130. Stefanis L. α-Synuclein in Parkinson's disease. Cold Spring Harb Perspect Med. 2012;2(2): a009399.
131. Braak H, Rüb U, Gai WP, Del Tredici K. Idiopathic Parkinson's disease: possible routes by which vulnerable neuronal types may be subject to neuroinvasion by an unknown pathogen. J Neural Transm (Vienna). 2003;110(5):517–36.
132. Paillusson S, Clairembault T, Biraud M, Neunlist M, Derkinderen P. Activity-dependent secretion of alpha-synuclein by enteric neurons. J Neurochem. 2013;125(4):512–7.
133. Holmqvist S, Chutna O, Bousset L, Aldrin-Kirk P, Li W, Bjorklund T, et al. Direct evidence of Parkinson pathology spread from the gastrointestinal tract to the brain in rats. Acta Neuropathol. 2014;128(6):805–20.
134. Chen SG, Stribinskis V, Rane MJ, Demuth DR, Gozal E, Roberts AM, et al. Exposure to the functional bacterial amyloid protein curli enhances alpha-synuclein aggregation in aged Fischer 344 rats and Caenorhabditis elegans. Sci Rep. 2016;6:34477.
135. Liu B, Fang F, Pedersen NL, Tillander A, Ludvigsson JF, Ekbom A, et al. Vagotomy and Parkinson disease: a Swedish register-based matched-cohort study. Neurology. 2017;88(21): 1996–2002.
136. Scheperjans F, Aho V, Pereira PA, Koskinen K, Paulin L, Pekkonen E, et al. Gut microbiota are related to Parkinson's disease and clinical phenotype. Mov Disord. 2015;30(3):350–8.
137. Keshavarzian A, Green SJ, Engen PA, Voigt RM, Naqib A, Forsyth CB, et al. Colonic bacterial composition in Parkinson's disease. Mov Disord. 2015;30(10):1351–60.
138. Baldini F, Hertel J, Sandt E, Thinnes CC, Neuberger-Castillo L, Pavelka L, et al. Parkinson's disease-associated alterations of the gut microbiome predict disease-relevant changes in metabolic functions. BMC Biol. 2020;18(1):1–21.
139. Perez-Pardo P, Kliest T, Dodiya HB, Broersen LM, Garssen J, Keshavarzian A, et al. The gut-brain axis in Parkinson's disease: possibilities for food-based therapies. Eur J Pharmacol. 2017;817:86–95.
140. Barichella M, Pacchetti C, Bolliri C, Cassani E, Iorio L, Pusani C, et al. Probiotics and prebiotic fiber for constipation associated with Parkinson disease: an RCT. Neurology. 2016;87(12):1274–80.

141. Berding K, Cryan JF. Microbiota-targeted interventions for mental health. Curr Opin Psychiatry. 2022;35(1):3–9.
142. Suez J, Zmora N, Segal E, Elinav E. The pros, cons, and many unknowns of probiotics. Nat Med. 2019;25(5):716–29.
143. Cenit MC, Sanz Y, Codoñer-Franch P. Influence of gut microbiota on neuropsychiatric disorders. World J Gastroenterol. 2017;23(30):5486–98.
144. Desai V, Kozyrskyj AL, Lau S, Sanni O, Dennett L, Walter J, et al. Effectiveness of probiotic, prebiotic, and synbiotic supplementation to improve perinatal mental health in mothers: a systematic review and meta-analysis. Front Psych. 2021;12:622181.
145. Ng QX, Soh AYS, Venkatanarayanan N, Ho CYX, Lim DY, Yeo WS. A systematic review of the effect of probiotic supplementation on schizophrenia symptoms. Neuropsychobiology. 2019;78(1):1–6.
146. Huang R, Wang K, Hu J. Effect of probiotics on depression: a systematic review and meta-analysis of randomized controlled trials. Nutrients. 2016;8(8):483.
147. Depeint F, Tzortzis G, Vulevic J, I'Anson K, Gibson GR. Prebiotic evaluation of a novel galactooligosaccharide mixture produced by the enzymatic activity of Bifidobacterium bifidum NCIMB 41171, in healthy humans: a randomized, double-blind, crossover, placebo-controlled intervention study. Am J Clin Nutr. 2008;87(3):785–91.
148. Vulevic J, Juric A, Walton GE, Claus SP, Tzortzis G, Toward RE, et al. Influence of galacto-oligosaccharide mixture (B-GOS) on gut microbiota, immune parameters and metabonomics in elderly persons. Br J Nutr. 2015;114(4):586–95.
149. Nyangale EP, Farmer S, Keller D, Chernoff D, Gibson GR. Effect of prebiotics on the fecal microbiota of elderly volunteers after dietary supplementation of Bacillus coagulans GBI-30, 6086. Anaerobe. 2014;30:75–81.
150. Bakken JS, Borody T, Brandt LJ, Brill JV, Demarco DC, Franzos MA, et al. Treating clostridium difficile infection with fecal microbiota transplantation. Clin Gastroenterol Hepatol. 2011;9(12):1044–9.
151. Kang DW, Adams JB, Gregory AC, Borody T, Chittick L, Fasano A, et al. Microbiota transfer therapy alters gut ecosystem and improves gastrointestinal and autism symptoms: an open-label study. Microbiome. 2017;5(1):10.

# Inflammation-Mediated Responses in the Development of Neurodegenerative Diseases

3

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**Abstract**

Since its first description over a century ago, neurodegenerative diseases (NDDs) have impaired the lives of millions of people worldwide. As one of the major threats to human health, NDDs are characterized by progressive loss of neuronal structure and function, leading to the impaired function of the CNS. While the precise mechanisms underlying the emergence of NDDs remains elusive, association of neuroinflammation with the emergence of NDDs has been suggested. The immune system is tightly controlled to maintain homeostatic milieu and failure in doing so has been shown catastrophic. Here, we review current concepts on the cellular and molecular drivers responsible in the induction of neuroinflammation and how such event further promotes neuronal damage leading to neurodegeneration. Experimental data generated from cell culture and animal studies, gross and molecular pathologies of human CNS samples, and genome-wide association study are discussed to provide deeper insights into the mechanistic details of neuroinflammation and its roles in the emergence of NDDs.

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**Keywords**

Immune system · Pro-inflammatory · Neuroinflammation · Neurodegeneration · Alzheimer's disease · Parkinson's disease · Amyotrophic lateral sclerosis · Multiple sclerosis

## 3.1 Introduction

Neurodegenerative diseases (NDDs) have been bringing significant impacts on some related aspects. Not only giving a challenge for health care system, but also bringing impacts on economic situation. For example, in 2018, almost 40 million people globally were diagnosed with Alzheimer's disease (AD). In economic point of view, based on a report released by an authorized body in 2015, the global cost expended for tackling AD was approximately US$818 billion and was projected that this expenditure would increase to US$ 2 trillion in 2030 [1, 2]. It is noteworthy that this number is only associated with AD-related cost without taking into account the cost for the other neurodegenerative diseases.

Other surprising data come from a report focusing on the global aging issues released by the United Nations in 2015. In that report, in 2050, the number of people aged 60 years and older globally could reach approximately 2.1 billion [3]. As this group of age is the most vulnerable group suffering from the NDDs, this number with its increment is threatening our health care system and economic aspects if no significant efforts are achieved in the near future for providing a good therapeutic treatment for NDDs.

It is believed that these figures may continue to increase as no treatment gives satisfying outcomes for curing the NDDs recently. Within 10 years, between 2002 and 2012, only one new anti-AD drug was approved by the US Food and Drug Administration after reviewing 214 candidates that had entered clinical trials [4]. This is even worse if we are looking at the fate of the other NDDs, such as amyotrophic lateral sclerosis and Huntington's disease [1]. Some challenges for developing new drugs for treating NDDs exist from a big hurdle faced by the compounds to cross the blood-brain barrier to get into the brain [5], no animal model that is closely related to the NDD characteristic as seen in humans [6], to the high amount of fund that must be expended [7].

To tackle those challenges, a strong collaboration among all related players (multidisciplinary academics, industry, public, and government) is a critical need. Given that the efforts for discovering and developing drugs could not be achieved in a short-time setting, this collaboration could ensure the sustainability of the efforts. As described above, a large amount of money that may have to be spent could also be overcome by a good collaboration among the related parties [1]. A comprehensive and holistic approach should also be taken into account as NDDs are not only about curative aspects, but also associated with other aspects, including the promotive, preventive, and palliative aspects. At this point, the integrative and collaborative actions are, once again, pivotal.

Multifactorial events have been known to take part in the pathogenesis of NDDs. This could be like a double-edged sword. In one edge, multifactorial-related disorders could be beneficial in terms of providing various targets that could be explored in the effort of drug discovery. However, on the other edge, these disorders bring difficulty to be treated due to the complex pathophysiological aspects involved in the pathogenicity of the disorders.

Here, we focus on the aspect of NDDs linked to their interrelation with inflammatory events. It has been known that inflammation is one of the key events involved fundamentally in the emergence and development of the NDDs. An in-depth understanding regarding this aspect should provide an insight for the purpose of seeking treatment for NDDs. Also, we describe various models that can be utilized for doing NDD studies.

## 3.2 Hallmarks of Neurodegenerative Diseases

NDDs are still counted as fatal diseases, although many advances have been achieved in terms of their treatment. NDDs mainly attack the central nervous system leading to detrimental consequences on the synaptic network and death of neurons. Despite many NDD risk factors having been elucidated, aging is apparently a critical factor for neurodegenerative events [8, 9]. The elderly typically display perturbation in their cytokine expression that could be detrimental as this may lead to an imbalance between the higher expression of pro-inflammatory cytokines and anti-inflammatory cytokines. This could result in the development of a condition called "inflammaging" which is characterized by a chronic low-grade inflammation [10].

The human body has developed a delicate inflammatory system to respond to either the attack of various noxious biological/chemical materials or the presence of tissue injuries. A complex interrelationship between the inflamed cells and the inflammatory factors, such as cytokines, plays a major role in determining the state of inflammation. In principle, inflammatory events should be balanced between pathological and physiological modulation. This means that once the inflammatory causes have been removed, the resolution of the inflammation should be achieved. However, in some conditions, such as in NDDs and in inflammaging, the inflammation cannot be resolved easily as in a chronic state of inflammation certain modifications have altered the way the immune system acts leading to an unachievable inflammatory resolution [11].

The relationship between aging brains and NDDs brings several interesting points of view. For example, it is proposed that neurodegenerative processes are linked inherently to the normal brain aging as it is difficult to seek aging brains with a good or normal function. Therefore, an interesting point of view defines NDDs as accelerated aging processes that may be caused by a complex network between many factors, such as genetics and environmental factors. However, this view has a drawback, especially in capturing the underlying mechanisms by which aging is linked to neurodegenerative diseases [9].

Overall, although no fixed definition has come into a consensus from the experts regarding NDDs, in some aspects, some agreements have been made. It is understandable that a proper definition of NDDs is a necessity in the effort of tackling the impact of NDDs.

The abnormal accumulation, folding, and aggregation of a specific protein, e.g., α-synuclein, tau, and β-amyloid (Aβ), in the central nervous system are the key contributors and the hallmarks for the pathogenesis of NDDs, such as in Alzheimer's disease (AD), Parkinson's disease (PD), Huntington's disease (HD), frontotemporal dementia (FTD), and spinocerebellar ataxia type 1. Although many advanced points have been achieved recently regarding some aspects involved in the pathogenicity of the specific disease-driving proteins, the mechanisms used by those proteins to facilitate the emergence and development of the NDDs are still unclear [1].

Another hallmark of NDDs could be related to the spread of the pathogenic proteins. It has been demonstrated that abnormal protein aggregates could spread throughout the brain [12]. Firstly, an aggregate of proteins somehow experiences an abnormal conformation. Further, these initial abnormal aggregates become a trigger for the other proteins inducing the conversion of their nature from normal to pathogenic proteins. One evidence supporting this interesting phenomenon is given by a study observing the effect of pathogenic conformation of tau protein seeded in the other models expressing the tau protein. This study found that the seeded abnormal tau acted as inducer to other in vitro models expressing tau proteins, turning tau proteins in these models into abnormal aggregates [13].

Moreover, some studies reported that the inoculation of the pathogenic tau protein into mutant human tau protein-expressing mice brain caused an induction of tau abnormalities as also seen in individuals suffering from tauopathies [14, 15]. At this point, a question raises regarding how a single pathogenic protein can generate different phenotypic characteristics of the disease. Other groups confirmed that neurons developed from induced pluripotent stem cells expressing mutations associated with tauopathy experienced several neurodegeneration-related events, including alteration of the transport mechanism of the mitochondria, change in either the splicing or the distribution of tau protein preceding the event of tau aggregation, and change in the maturation of the cells [16]. Recently, no satisfying explanation has been provided to answer some attractive questions, e.g., whether all tau assemblies experience further spreading or only a specific tau assembly and whether the pathogenicity of the abnormal tau protein affects all types of cells or it is toxic only for specific cells [1].

Inflammation is another key contributor of NDD pathogenesis. Although the connection between the aggregation of the pathogenic proteins and the emergence of the NDDs has been accepted to explain the etiology of the NDDs, recently some changes in defining the etiological aspects of the NDDs have been proposed. For example, it has been known that Aβ aggregation has been linked closely to the pathogenesis of AD. However, some studies have proposed another hypothesis which states that inflammation is the main causative factor of neurodegeneration [17, 18]. It has also been indicated that before the emergence of the neurodegenerative events, various inflammatory pathways are activated resulting in the

enhancement of the inflammatory cascades. This intriguing hypothesis should provide alternative for NDDs' pharmacological intervention by targeting the pro-inflammatory factors, such as cytokines, and inflammation-related cascades [19].

## 3.3 The Role of Inflammation in the Development of Neurodegeneration

In the past years, increasing awareness on the important role of the innate and adaptive immune systems in the emergence of neurodegeneration has been one of the critical aspects in the study of neuroimmunology [20]. Many cells of the innate and adaptive arms of immunity are found throughout the human body. Essentially, these cells and their products define the existence of the immune system which possesses various pivotal roles: ranging from maintaining tissue homeostasis and repairing tissue injuries to host defense against pathogenic invaders [20, 21]. In the central nervous system (CNS), microglial cells are the main immune cells playing duties to ensure the physiological homeostasis takes place. Under normal condition, microglia produce various factors which play roles in anti-inflammatory and neurotrophic events in the CNS [22]. However, this condition changes when there is an invasion of pathogens leading to the production of inflammatory factors by microglia to counter the invasion [23].

Once the pathogens are eradicated and tissues are repaired, inflammatory response should be switched off. In fact, in several cases, the resolution of inflammation is disturbed [24]. Although inflammation is a normal mechanism and is involved in many beneficial effects, uncontrolled inflammation is a danger for the environment homeostasis [24]. The deposited pathogenic proteins are perceived by the immune system as a threat thereby activating the inflammatory processes. Unfortunately, in the case of NDDs, the accumulation of the proteins persists, and this could lead to the uncontrolled inflammatory responses.

The deposited pathogenic proteins are sensed by the immune cells, such as macrophages, via pattern recognition receptors, such as Toll-like receptors (TLRs), which are responsible for the recognition of pathogen-associated molecular patterns [25]. This interaction results in the recruitment of more immune cells, initiates adaptive immunity, and promotes the production of antimicrobial factors. These events are mediated by various cytokines released by the cells (i.e., TNF-α and IL-1β), chemokines (i.e., MCP-1), and other molecules (i.e., iNOS, ROS). In the context of NDDs, as the frontline defense in the CNS, microglia appear to be the major cells responsible for generating and maintaining inflammatory response because other immune cells may have a difficulty in infiltrating the CNS because of the existence of the blood-brain barrier (BBB). For a simplified reference, a schematic figure depicting proposed general inflammatory events involved in the development of NDDs is provided (Fig. 3.1).

Among all systems available in the human body, the CNS is previously considered to be immune-privileged [26], suggesting that the presence of non-self-antigens in the CNS is unable to induce adaptive immune responses. This was primarily

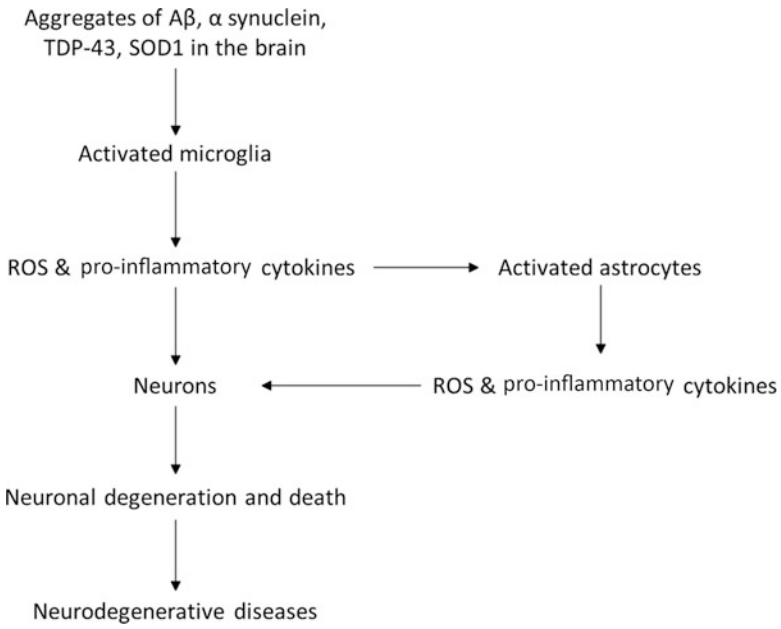


**Fig. 3.1** Proposed general inflammatory events involved in the development of neurodegenerative diseases

discussed by Sir Peter Medawar, a Noble Prize winner, over 70 years ago [26]. However, recent evidence indicated that the concept of immune privilege is not an unequivocal notion since immune responses have been shown to play a critical role in the mitigation of infection in the CNS [27–29]. In response to this, increasing attention to investigate close connection between inflammatory responses and the emergence of NDDs has been reported [30, 31]. A number of evidence strengthen the involvement of inflammatory events in the course of the diseases [20, 31]. A comprehensive understanding on this issue, including the specific role of the immune system in mediating inflammation-related neurodegenerative diseases, will in turn refuel the efforts to develop drugs used for treating the diseases.

Since its first description over a century ago, NDDs have been mainly studied based on changes appearing in the gross anatomy of the neurons and related organs as well as changes in the integrity of neurons leading to neuronal loss [32]. However, recent data generated from genetic, histopathological, and molecular studies revealed the involvement of immune dysregulation phenotypes such as alteration in the cytokine production and signaling, changes in the proliferation of migration of immune cells, and improper modification of phagocytosis behavior, in the development of NDDs [32]. These immunological changes have been presently noted as important players in the onset and progression of neurodegenerative diseases [20, 32].

How can inflammation of the CNS provoke neurodegeneration? While this is a simple yet important question in the study of NDDs, it proves to be difficult to address. Preliminary findings from in vitro and in vivo animal studies suggested the role of neuroinflammation in the development of NDDs [32–34]. Since this notion is fairly supported by limited human studies, hallmarks of neuroinflammation and the intricate relationship between neuroinflammation and NDDs remain difficult to define in a general term [35]. Phenotypical events such as intense glial responses and impairment of the blood-brain barrier leading to extravagant infiltration of the blood-circulating lymphocytes and monocyte-derived macrophages into the CNS parenchyma were observed in the cases of multiple sclerosis (MS). However, in contrast to the unambiguous role of neuroinflammation in the development of MS, such events are not typically observed in other neurodegenerative diseases such as AD, PD, and amyotrophic lateral sclerosis (ALS). Instead of the typified massive infiltration of systemic immune cells into the CNS, neuroinflammation observed in AD, PD, and ALS is characterized by increasing activity of astrocytes and microglia with the presence of inflammatory mediators in the CNS parenchyma at a low to moderate concentration [35].

Neuroinflammation is a distressing manifestation of immune dysregulation and the role of such event in the development of NDDs, including AD, PD, ALS, and MS, at different stages of the diseases has been increasingly appreciated [20, 32]. Signatures of the innate and adaptive immune responses leading to the induction of neuroinflammation have been observed and characterized [20], emphasizing the notion of inflammation-mediated neurodegeneration. At present, the roles of several immune cells (Table 3.1) in the development of NDDs have been suggested. These cells are predominantly involved in the induction of neuroinflammation, leading to neuronal cell death and ultimately neurodegeneration.

## 3.4 The Role of Neuroinflammation in the Pathogenesis of Neurodegenerative Diseases

Although different pathological mechanisms underlie many NDDs, the formation of pathological insoluble aggregates formed from a specific protein deposition in the CNS seems to be the shared pathological identity among the diseases (Table 3.2) [37, 38]. The accumulation of the pathologic proteins can result from two main factors, i.e., the excessive production of the proteins and the impairment of the clearance mechanism of proteins where both factors can work either independently or together. Intriguingly, growing evidence shows that abnormal proteins can be transmitted among cells through pathways that are being investigated intensively.

Neurons usually become the preferred site for experiencing dysfunctionalities following the pathogenic protein deposition. Several characteristics support the vulnerability of a neuron including its long axon which can reach a meter or more to get into contact with the adjacent neurons, the inability to undergo mitosis (postmitotic), the inability to have a proper regeneration process once it is degenerated, and its complex synaptic connection [38]. Neuronal damage will

**Table 3.1** Immune cells involved in neuroinflammation and neurodegeneration

| Types | Immune cells | Physiological function | Roles in neuroinflammation and neurodegeneration |
|---|---|---|---|
| Innate immune cells | Astrocytes | Astrocytes play a major role in the formation and maintenance of synapse function, providing a physiological support for neurons. In addition, astrocytes also provide support during synaptic pruning by phagocytes [20] and maintain the integrity of the BBB and the homeostatic concentration of neurotransmitters and ions at the extracellular region [36] | There are two different phenotypes of astrocytes: A1 (inflammation-induced) and A2 (ischemia-induced). In the event of immune dysregulation, inflamed astrocytes, particularly A2, were present. These cells have been shown to produce pro-inflammatory cytokines and chemokines, which will induce the recruitment of monocytes into the CNS, activate more astrocytes, and further induce the inflammation in the CNS [20]. In addition, in the event of infection or the release of danger-associated molecular patterns (DAMPs), pathogen recognition receptors (PRRs) such as TLR will mediate the activation of microglia which are necessary for the induction of A1 astrocytes in a manner dependent on the combination of IL-1α, TNF, and complement C1q. In the CNS tissues obtained from MS, PD, AD, and ALS patients, A1 astrocytes expressing downstream complement C3 were found, suggesting their important roles in the induction of inflammation-mediated neurodegeneration [20] |
| Innate immune cells | Microglia (microglial cells) | Microglial cells serve as phagocytic cells, providing a first line of defense against pathogens in the CNS. Non-self-antigens produced by pathogens or injured cells are recognized via various TLRs expressed on the surface of microglia, leading to the expression of pro-inflammatory mediators followed by subsequent phagocytosis by the corresponding microglia. Prolonged expression of pro- | Upon exposure to inflammatory insults, microglial cells are rapidly activated and subsequently migrated to the source of insults. Activated microglia are able to express surface molecules such as CD14 and major histocompatibility complex (MHC), allowing them to interact with T cells [21]. In the pathological events leading to severe inflammatory condition, microglial cells retain |

(continued)

**Table 3.1** (continued)

| Types | Immune cells | Physiological function | Roles in neuroinflammation and neurodegeneration |
|---|---|---|---|
| | | inflammatory cytokines will promote subsequent recruitment of other microglia into the sites of infection/injury [21]. In addition to its immunological function, microglia also serve the purpose to promote new synapse formation that leads to the differentiation and proliferation of neurons [21] | amoeboid M1-like phenotypes characterized by increased expression of pro-inflammatory cytokines, inducible nitric oxide synthase (iNOS), and reactive oxygen species (ROS) [21]. Prolonged expression of such pro-inflammatory cytokines in the CNS can induce neuroinflammation and neuronal cell death, leading to neurodegeneration [23] |
| Adaptive immune cells | CD4+ T helper lymphocytes | As one type of lymphocytes, CD4+ T cell is an important player in the activation of adaptive immune responses. Its main function is mainly related to the orchestration of adaptive immune cells, including the initiation of B cell-mediated production of antibodies. CD4+ T cells are classified into several cell types including T-helper 1 (Th1), Th2, Th17, and Tregs. Proper levels of Th1 and Th2 and the balance between these two cell types determine the healthy condition of the CNS environment [21]. In addition, production of a wide range of cytokines by Th17, including IL-17A, IL-17F, IL-22, and IL-21, is known to mediate host immune responses and the activation of Treg is essential in the maintenance of immune tolerance in the CNS [21] | Elevated levels of pro-inflammatory cytokines, including IFN-γ and TNF-α (due to hyperactivation of Th1 cells) as well as IL-17 and IL-22 (due to Th17 overactivation), have been suggested to play a role in the neuroinflammation-mediated neurodegeneration [21] |
| Adaptive immune cells | CD8+ T cytotoxic lymphocytes | In addition to CD4+ T cells, CD8+ T cells (cytotoxic T cells) play a vital role in the recognition of pathogen-infected cells to maintain the cellular function of adaptive immune responses. This has been suggested as an important value in the maintenance of homeostasis in the CNS [21] | The exact role of CD8+ T cells in the development of neuroinflammation remains unclear. However, current data implicated its role in the pathophysiology of NDDs, predominantly MS [21] |

**Table 3.2** Neurodegenerative diseases and the accumulation of their related abnormal proteins

| Neurodegenerative diseases | Protein deposition |
|---|---|
| Alzheimer's disease (AD) | Amyloid beta (Aβ), tau |
| Parkinson's disease (PD) | α-Synuclein |
| Huntington's disease (HD) | Huntingtin |
| Amyotrophic lateral sclerosis (ALS) | TDP-43, SOD1 |
| Frontotemporal dementia (FTD) | Tau, TDP-43 |
| Prion disease | PrP |

ultimately affect physiological processes occurring in glial cells and synapses leading to the impairment of their connection system [39]. However, a growing body of evidence shows that the accumulation of the proteins can also take place within glial cells, such as astrocytes and oligodendrocytes [40].

In this section, we will only focus on how inflammation could play a significant role in the pathological mechanism of AD, PD, ALS, and MS.

### 3.4.1 Alzheimer's Disease

Since its first identification more than a century ago, many important milestones related to the formation and progression of Alzheimer's disease have been reached. However, there are still many unknown aspects of this disease and there is still no satisfying therapy for the patients who are suffering from this NDD.

It has been known that the culprits responsible for the occurrence of Alzheimer's disease are amyloid plaques and neurofibrillary tangles (NFTs) [41]. While the amyloid plaques are formed from the aggregates of Aβ extracellularly, NFTs are developed intracellularly following the hyperphosphorylation of tau proteins responsible for stabilizing neuronal microtubules [41].

Aβ is formed from the cleavage of the amyloid precursor protein (APP) which is a type I transmembrane protein with N-terminus located in the ectodomain and C-terminus in the cytosol. It has three isoforms which differ from the number of their amino acid residues. The 695-amino acid isoform is abundantly found in the neuron, while the others, the 751- and 770-amino acid isoforms, are mostly expressed systemically. The Aβ domain contains 40–43 amino acid residues and is located in the middle of the APP [41]. The gene that codes the APP is mapped in chromosome 21q21.3, while the microtubule-associated protein tau is encoded by a *MAPT* gene located in chromosome 17q21 [39]. As APP, several isoforms are also detected for the protein encoded by *MAPT* gene.

The proteolytic cleavage of the APP occurs via either amyloidogenic or non-amyloidogenic pathways. Both pathways utilize secretases as the catalyst. The secretase enzymes are divided into three types, i.e., α-, β-, and γ-secretases. In the non-amyloidogenic pathway, the α-secretase cleaves the APP at amino acid 17 within the Aβ domain where this action inhibits the formation of Aβ. This action produces two products, i.e., APP-α ectodomain and APP-CTF83 (membrane-bound

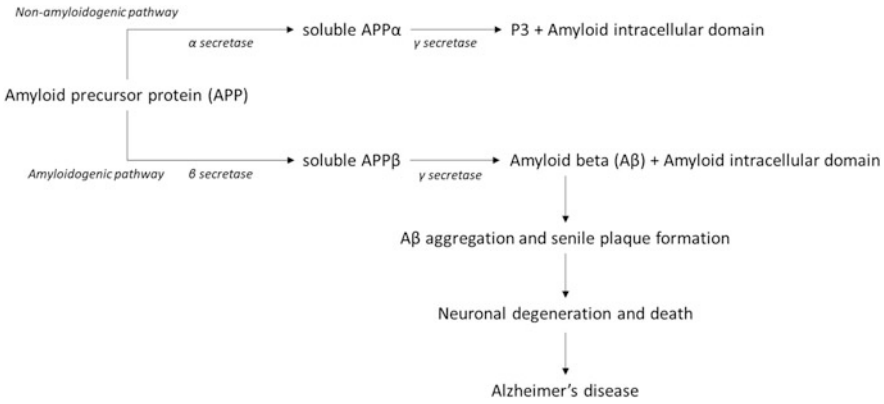


**Fig. 3.2** Biogenesis of amyloid beta and its proposed impact on the development of Alzheimer's diseases

C-terminal fragment containing 83 amino acids of APP). The latter product is further cleaved by γ-secretase to generate the P3 fragment and AICD [41–43].

In the amyloidogenic pathway, the β-secretase cleaves the APP generating two products, i.e., APP-CTF99 (membrane-bound C-terminal fragment containing 99 amino acids of APP) and APP-β. The former domain undergoes further cleavage by the γ-secretase generating two other domains, i.e., Aβ and APP intracellular domain (AICD). There are two Aβ peptides produced by γ-secretase, which are soluble $A\beta_{1-40}$ and insoluble $A\beta_{1-42}$ [42]. Although the latter is the minor product generated from this metabolism process, it is the main composition found in amyloid plaque compared to the former peptide. A simplified explanation on the biogenesis of amyloid beta and its proposed impact on the development of Alzheimer's diseases is presented in Fig. 3.2.

During the amyloidogenic processing of APP, $A\beta_{1-42}$ is secreted as a monomer. The monomeric form of $A\beta_{1-42}$ has a propensity to aggregate into oligomers which further form fibrils. The ultimate form of this aggregation process is the formation of amyloid plaques which have been identified as the hallmarks of AD [41, 42]. The plaques are linked to many harmful effects, especially on neuronal activity where the plaques can cause disruption of synaptic function, intracellular signaling, inflammatory cascade, and cytoskeleton activity.

Several studies reported that $A\beta_{1-42}$ peptides were released in a physiological level to assist several processes such as the release of neurotransmitter, the removal of excessive metal ions, and the cellular protection on oxidative stress [44–46]. However, in Alzheimer's disease, this homeostasis is disturbed. Overproduction of $A\beta_{1-42}$, excessive supply of $A\beta_{1-42}$ from the systemic circulation, and impairment of $A\beta_{1-42}$ clearance from the brain are the main factors causing the toxic accumulation of $A\beta_{1-42}$ in the brain [42]. In this regard, the role of the receptor for advanced glycation end products (RAGE) in mediating the influx of $A\beta_{1-42}$ into the brain and low-density lipoprotein-related protein (LRP) receptor 1 (LRP1) which facilitates

the efflux of the peptides from the brain is an interesting topic to be further elucidated [47–50]. Related to this, mutations occurring in the APP and γ-secretase have been found to have contributions to familial AD [51]. In addition, the clearance of Aβ may also be carried out by a mechanism which involves the interaction between microglia and apolipoprotein E (apoE) [52].

In addition to oxidative stress and tau hyperphosphorylation, inflammation plays an important role in assisting AD progression. The excessive accumulation of amyloid plaques and the presence of NFTs can stimulate the production of pro-inflammatory cytokines, chemokines, and radical oxygen species from associated immune cells, such as microglia [53, 54]. Specifically, the level of IL-1β, IL-6, MHC class II, COX-2, MCP-1, TNF-α, IL-1α, CXCR2, CCR3, CCR5, and TGF-β is elevated in the brains of patients suffering from AD [53, 55, 56]. The uncontrolled production of those molecules is associated with neuronal death.

In the brain, Aβ accumulation is sensed by microglia through TLRs and other sensors, such as CD14 and MD2, expressed in microglia [25]. TLR4 is proposed as the specific TLRs involved in this sensing process. The activation of TLRs expressed in glial cells is followed by the activation of the subsequent transcription factors that will upregulate the inflammatory gene expression [57, 58].

NOD-like receptors (NLRs) are the next receptor involved in the sensing system of Aβ [25]. The presence of this senile protein induces a member of NLRs called NALP3 expressed in microglia which is responsible for the activation of various signaling proteins implicated in the occurrence of apoptosis and maturation of pro-inflammatory cytokines, e.g., IL-1β and IL-18 [25, 59].

In addition to its role in mediating influx of the Aβ, RAGE is identified as another sensing system expressed in various cells, including microglia, astrocytes, vascular endothelial cell, and neurons [25]. This receptor was initially found as a receptor for advanced glycation end products (AGEs). However, several studies confirmed that this cell surface receptor also has an affinity to bind Aβ [60]. The Aβ-RAGE complex activates microglia which is followed by the production of pro-inflammatory cytokines, such as TNF-α, IL-1β, and IL-6 through the activation of caspases and signal-dependent transcription factors such as NF-kB and AP-1 [25]. These cytokines may be responsible for the neuronal apoptotic mechanism upon their direct binding to the neuron.

Reciprocal communication between microglia and astrocytes also plays a certain role in inducing inflammation in the course of AD. The cytokines released by astrocytes may cause further activation of microglia and vice versa. Intriguingly, it has been found that the APP, γ-secretase, and β-secretase are found to express NF-κB in their promoter region [61]. Once the NF-κB sites are occupied by the cytokines, subsequent upregulation of the APP and the secretases occurs in neurons and this is linked to further induction of Aβ secretion. Ultimately, the excessive production of the Aβ from the neuron is followed by the activation of microglia, and this will lead to the aggravation of microglia-mediated inflammation in AD [25, 62].

To sum up, upon the production of Aβ aggregates and plaques, sensing systems in microglia are activated, mainly TLRs and RAGE. The activation of these systems is followed by the release of some related pro-inflammatory factors, such as cytokines,

chemokines, and ROS which is mediated by NF-κB and AP-1 signaling pathways [25]. Next, these factors induce astrocytes and neurons so that the pro-inflammatory signal is exaggerated leading to neurotoxic effects where the neuron suffers from either apoptosis or necrosis events [63, 64]. ATP released from the death neurons is then taken up by microglia via the P2X7 receptor, a purinergic receptor, which is involved in the aggravation of inflammatory responses [65]. Several studies suggested that cholinergic neurons were found as the most vulnerable target to undergo neurotoxicity after an Aβ-induced inflammatory course, while other neurons, such as GABAergic and glutaminergic, may be also susceptible [25, 66, 67].

### 3.4.2 Parkinson's Disease

As in AD, the pathogenesis of PD is linked to the accumulation of a misfolded neuronal protein. The protein, recognized as α-synuclein, is a member of the synuclein family consisting of two other synucleins, β- and γ-synuclein [68]. Of those synucleins, α-synuclein has been identified as the primary culprit in the development of PD as α-synuclein is the major component found inside the intraneuronal inclusions called Lewy bodies which are responsible for the pathogenesis of PD [68, 69].

α-Synuclein consists of 140 amino acids and is encoded by the SNCA gene located in 4q21 [39, 70]. Natively formed in the unfolded monomer form in the cytoplasm, α-synuclein is further converted into misfolded α-helical secondary protein after having contact with lipid components in the lipid membranes [69–71]. The product has a propensity to form aggregates with other monomers and is linked to neurotoxic effects [70].

Posttranslational modification processes, including phosphorylation, nitration, and ubiquitination, are also implicated in the neurotoxic property of the protein [70–74]. It has been proposed that posttranslational modification of α-synuclein positively correlates with the formation of Lewy bodies [71]. The modification may increase the insolubility, induce aggregation, change the localization, inhibit clearance, and promote the neurotoxic effect of α-synuclein [70, 73–75]. Phosphorylation of α-synuclein occurs mostly in serine residues, while ubiquitination and nitration predominantly take place in lysine and tyrosine residues, respectively [70, 76, 77].

To date, the physiological roles of α-synuclein are still unclear. The soluble and membrane-bound form of α-synuclein is found to be balanced in physiological conditions [68]. One study proposed that α-synuclein was involved in the release of neurotransmitters from the presynaptic terminal in dopaminergic networks [70]. Accordingly, the accumulation of this protein may lead to the emergence of dopaminergic neuron dysfunctionalities located mainly in the substantia nigra of the brain resulting in the emergence of several specific PD symptoms which are either related to motor disturbances (tremor, rigidity, bradykinesia) and non-motor-related

symptoms, e.g., deficits in olfactory and cognitive functions, disturbances in autonomic nervous system, and sleep disorders [25].

Unlike amyloid beta, α-synuclein tends to accumulate intracellularly. However, a study suggested that α-synuclein could be secreted out of the neuron and formed α-synuclein aggregates [78]. After this accumulation, several pathogenic effects occurred such as neuroinflammation, neurodegeneration, and cell death [79]. Interestingly, neurodegeneration may also be caused by the failure of α-synuclein in playing its physiological roles [80].

α-Synuclein accumulation-induced inflammation begins when microglia sense the deposition of the protein. As microglia do not exhibit a specific sensing system to sense the accumulation of α-synuclein [25], the sensing process occurs when the protein directly binds to microglia to begin inflammatory cascades. Upon the uptake of α-synuclein by microglia, NF-κB is activated which is followed by the increased production of several pro-inflammatory factors such as cytokines (TNF-α and IL-1β), radical oxygen species through the activation of NADPH oxidase, and NO via the action of inducible nitric oxide synthase (iNOS) [25]. These factors act directly on dopaminergic neurons as the main target in PD. In addition, these factors also activate astrocytes. The activated astrocytes also release pro-inflammatory factors which attack the dopaminergic neurons amplifying the attack of the factors produced by microglia [25]. Finally, the products produced by both microglia and astrocytes as a response for α-synuclein aggregation induce neurotoxicity [25].

Several environmental toxins have been found to produce a neurotoxic effect on dopaminergic neurons. 1-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) is a prodrug which is converted in glial cells into its metabolite named 1-methyl-4-phenylpyridinium (MPP+) [81]. This metabolite is recognized as a neurotoxin causing permanent symptoms of PD by destroying dopaminergic neurons [25] via induction of mitochondrial damage-related oxidative stress.

Bacterial lipopolysaccharide (LPS) also displays an ability to induce neuronal death by inducing the release of inflammatory factors from nonneuronal cells, such as microglia [82]. Unlike the TLR-independent mechanism shown by α-synuclein, LPS can be sensed by microglia particularly via TLR4 [83]. By utilizing this mechanism, LPSs subsequently induce the release of pro-inflammatory factors mainly via the activation of NF-κB and finally cause loss of dopaminergic neurons in the substantia nigra [25].

### 3.4.3 Amyotrophic Lateral Sclerosis

This progressive NDD was first identified by the French neurologist, Jean Martin Charcot, in 1874. Although many important things have been revealed, the disease, also known as Lou Gehrig's disease, is still incurable.

In ALS, three major sites in the brain which are related to motor function initially malfunction, i.e., the brainstem, spinal cord, and motor cortex [25]. The dysfunctionalities of these motor neurons lead to the emergence of clinical features of ALS, e.g., muscle twitching (fasciculation), muscle loss, weakness, spasticity, and

some respiratory complications such as respiratory muscle and diaphragm paralysis, which are described as the main causes of ASL-related death [25].

As other progressive NDDs, the pathogenesis of ALS is also linked to the deposition of a pathological protein called transactive DNA-binding protein-43 (TDP-43) within the cytoplasmic ubiquitin inclusion in degenerating motor neurons [84]. The protein is encoded by the *tardbp* gene located in chromosome 1 [39, 84]. Although the abnormal TDP-43 deposition in abnormal intracellular ubiquitin inclusion is seen in the sporadic form of ALS, TDP-43 deposition is also observed in the familial form of ALS due to mutation occurring in the *tardbp gene* [85–88].

TDP-43 is a protein consisting of 414 amino acids [84]. Physiologically, TDP-43 does not form an inclusion as it is involved in several normal processes such as in the regulation of RNA splicing, stability, transcription, and translation [84, 89]. The formation of insoluble TDP-43 aggregates occurs when the protein mislocalizes in the cytoplasm [84].

Posttranslational modification is the critical step in emanating pathological TDP-43. Two processes in PTM that have been identified as the key processes involved in generating pathological TDP-43 are phosphorylation and ubiquitination [84]. Neumann et al. reported that either ubiquitinated or phosphorylated TDP-43 was found abundantly in ALS patients [88]. The abnormal accumulation of TDP-43 in neurons followed by dysfunctionalities of the neuron and glial cells is proposed as the primary pathogenesis of ALS [84]. In *Drosophila melanogaster*, the accumulation of TDP-43 is associated with a swollen axon leading to transmission disturbance in motor neurons and ultimately causes motion disability [90].

The accumulation of TDP-43 is exacerbated by the clearance inhibition of TDP-43 [84]. Two mechanisms have been identified as the primary clearance routes for pathological TDP-43, i.e., exocrine secretion and autophagy [91, 92]. Barmada et al. demonstrated that autophagy stimulation could diminish mislocalization of TDP-43 leading to the increase of neuronal survival after being deposited by TDP-43 [91].

Other genes are also responsible for the heritability of ALS including superoxide dismutase 1 (*SOD1*) [93, 94] and *FUS/TLS* (fused in sarcoma or translocation in liposarcoma) [95, 96]. These genes are implicated to the production of other neurotoxic proteins SOD1 and FUS/TLS, respectively, also found abundantly in ALS [25].

Following the deposition of TDP-43 and SOD1 aggregates, inflammatory reaction emerges. Like the previously described NDDs, microglia also utilize TLRs along with their co-receptor, CD14 (cluster of differentiation 14), as the sensing system towards the pathological aggregates formed. Subsequently, NF-κB and AP-1 signaling pathways are activated resulting in the release of pro-inflammatory-related factors, such as cytokines, chemokines, and ROS. Intriguingly, the abundant release of IL-1β and TNF-α causes neurotoxic effects in vitro only, while there is no satisfying evidence supporting the role of these cytokines in ALS in vivo as deletion of the genes coding those cytokines in animal models produces no significant inhibition in ALS progression [25].

Those inflammatory factors then attack the motor neurons and activate the astrocytes. In turn, the activated glial cells also release pro-inflammatory factors which are also involved in generating neurotoxic effects on the motor neurons. The activation of microglia could also occur when the dying motor neurons release ATP which is then sensed by the purinergic receptor P2X7 expressed on them [25, 65].

### 3.4.4 Multiple Sclerosis

Multiple sclerosis is a neurodegenerative disease characterized by inflammation and demyelination occurring in the CNS leading to damaging impacts on the motor, autonomic, cognitive, and visual systems of patients [97]. In an autoimmune disease, immune cells attack the myelin sheath leading to axonal dysfunction and ultimately neuronal degeneration. Intriguingly, although protein deposition is observed in MS as seen in the previously described NDD, it does not relate to the pathogenic characteristic [25].

There is no satisfying evidence supporting the inheritability of this disease, suspecting the role of environmental factors as the initiator for the emergence of MS. Microbial infection is suspected as the potent factor that could initiate MS as certain regions found in viruses and bacteria are known to express antigenic pathogen-associated proteins which display similarity to myelin basic protein (MBP) expressed in myelin sheath of the neuron [25].

The involvement of both innate and acquired immunity in MS pathology has been described. It has also been shown that APCs circulating outside the CNS also play a pivotal role in the progression of MS. Some antigen-presenting cells (APCs), such as dendritic cells and macrophages, including microglia in the CNS, mediate the recognition of the antigens by lymphocytes (Glass). The induction of Toll-like receptors (TLRs) expressed in microglia and astrocytes triggers NF-κB and AP-1 signaling pathway [25].

Following the myelin-derived antigen recognition, microglia release IL-6 and TGF-β, which are responsible for the induction of naïve T cells to differentiate into Th17 cells expressing retinoic acid receptor-related orphan receptor γt (RORγt) [98, 99]. The activated microglia release IL-23 inducing Th17 to secrete IL-17 and TNF-α [100]. This Th17 induction could also be stimulated by osteopontin which is released by the activated astrocytes [101]. These excessive secreted cytokines impair the myelin sheath resulting in axonal damage [25].

Furthermore, the activated astrocytes secrete BAFF (B cell-activating factor belonging to the TNF family) differentiating into plasma cells responsible for the production of anti-myelin antibodies [102]. In addition, the activated astrocytes and microglia release ROS and NO which are also involved in the impairment of the myelin sheath. Finally, the massive damage occurring in the myelin sheath could not be repaired by oligodendrocytes, the glial cells responsible for producing new myelin [25].

## 3.5 Model Systems Available to Study Inflammatory-Mediated Neurodegenerative Diseases

The development of in vitro and in vivo models for studying NDDs is still experiencing several challenges. Many studies reported their success in testing drug candidates using the current models, but later reported their failure to translate the results into clinical settings. No models can completely phenocopy human diseases, including NDDs [103]. While each model comes with its unique advantages, at the same time some aspects limit the ability of the model to mimic the development and progression of NDDs in humans.

### 3.5.1 Alzheimer's Disease

AD is mainly identified by two hallmarks, i.e., Aβ deposition leading to the formation of insoluble plaques and the formation of neurofibrillary tangles [41]. These pathogenic events lead to the progressive degeneration of the hippocampus and other related parts of the brain. Given these key events of AD, the ideal model for AD should facilitate the formation of Aβ and neurofibrillary tangles and should display processes involved from the beginning to the occurrence of neural degeneration resulting in behavioral changes.

According to these key pathogenic events, transgenic APP rodents could not be used to capture all events that occurred as these models only increase the formation of Aβ, but not for tau proteins, the major proteins involved in the formation of neurofibrillary tangles [103]. Consequently, the use of these mutants in studying cognitive and behavioral studies in AD is not recommended [104, 105].

Although animal models offer more complex anatomical and neurobiological systems compared to in vitro models, the use of animal models is exclusively limited in mimicking early-onset familial AD which is only found in approximately 5% of all AD cases [33, 103, 106]. The etiology of familial AD is associated with mutations occurring in the three main genes, i.e., amyloid precursor protein (APP), presenilin 1 (PSEN1), and presenilin 2 (PSEN2) [103, 106]. Conversely, the common type of AD, late-onset AD or sporadic AD, is experienced by patients who are over the age of 65 years. The major cause of this AD type is still unknown and is believed to be multifactorial, making the design for developing the appropriate model challenging as the genetically engineered rodents that have been developed are almost exclusively put as the model for the familial AD [33, 107].

Another limitation faced by most animal models, such as rodents, in mimicking AD is associated with the difficulty in forming senile plaques and neurofibrillary tangles [33]. As AD is not found naturally in rodents [108], the injection of Aβ into the animals is typically undertaken. Yet, although the models can be injected with Aβ and tau proteins, these molecules rarely develop into senile plaques and NFTs, respectively [33]. This may be caused by the fact that the formation of the plaques and tangles needs years which cannot be compared to the short life span of the rodents [105]. Furthermore, the accumulation of the proteins in genetically modified

mice remains enigmatic as the models cannot lead to extensive neuronal loss and behavioral changes as seen in AD patients [103].

As the closest animal having physiological similarity to humans, nonhuman primates are the promising models for deciphering the underlying mechanisms of AD and in testing drug candidates for AD treatment [109]. By having this similarity supported by their complex neuroanatomy as well as cognitive and motor functions, nonhuman primates play an important role in AD research [110]. However, several hurdles limit the use of nonhuman primates. Although Aβ and tau protein as well as the formation of the plaques and the tangles can be seen naturally in some aged nonhuman primates, the distribution of these proteins is not identical with those in humans [111]. For example, in Rhesus macaques, Aβs are deposited mainly in the limbic cortex and frontal area of the brain [109]. Only a small portion of Aβ is deposited in the hippocampal area, which is the main site of Aβ deposition in humans [109]. Another example could be seen in tau protein accumulation. As in Aβ, the accumulation of tau protein in humans is mainly detected in the hippocampal area, while in mouse lemurs, this protein is accumulated in the cerebral cortex [109]. Ethical and financial issues are also on the list of the drawbacks which ultimately affect the size of the animal samples, which is typically small [109].

In addition to rodent and nonhuman primate models, the use of other animal models is also utilized. These include *Drosophila melanogaster* [112], *Danio rerio* [113], and *Caenorhabditis elegans* [114]. They are used to gain information regarding the potential effects that can be generated by Aβ and tau proteins either on the cellular or organism level.

Two-dimensional (2D) in vitro models are also useful in the study of AD as these models bring several clear advantages, i.e., easy to conduct genetic modification and genome screening, shorter time needed to perform the experiments, ease of handling, and relatively cheap [115]. However, two-dimensional in vitro models are not the model of choice for studying the complexity of physiological systems as well as for looking deeper at the aspect of developmental biology [115]. These models are invaluable as they can provide insights into how neuronal dysfunction and cellular loss occur in AD [116].

PC12, HEK293, and SH-SY5Y cell lines are commonly used and they are easy to be transfected with Aβ and tau proteins [116]. In addition to immortalized cell lines, the use of primary cortical and hippocampal culture systems is also common [116]. Some groups have utilized 2D models developed from induced pluripotent stem cells (iPSC) [117–119]. Yagi et al. found that neurons that differentiated from iPSC isolated from fibroblasts in patients suffering from early-onset AD with PSEN1 and PSEN2 mutations showed higher production of Aβ42 compared to the healthy control group [119]. Moreover, Israel and coworkers reported that the administration of the β-secretase inhibitor on iPSC produced a reduction of the tau protein and GSK-3β, an enzyme mediating the effect of Aβ on the activation of tau protein [117].

Although 2D models have provided answers on some pathophysiological events occurring in AD, the models are not free of drawbacks. It has been observed that Aβ secreted within the models has insufficient levels to produce senile plaques [120]. It is also noteworthy that glial cells, such as microglia and astrocytes, play a significant

role in AD development [121, 122]. At this point, co-culture between 2D models of neuronal cells and glial cells is pivotal in the effort of providing more relevant models.

The drawbacks faced by the 2D models inspire researchers to generate and develop 3D models. Choi et al. developed the first 3D neuronal model which differentiated from an immortalized human neural stem cell line [123]. The model is powerful as it does not only differentiate into neuronal cells, but also into glial cells [123]. This 3D model not only secretes Aβ and tau proteins, but also facilitates the formation of senile plaques and neurofibrillary tangles, respectively [123].

Brain organoids are another powerful in vitro model in studying AD. This model improves the features that have been achieved by previous models. For example, vascularized human cortical organoids have been developed in some studies to overcome the lack of oxygen and nutrition faced by conventional brain organoids [124–127]. This model is an eminent model as it can also express some major blood-brain barrier markers, such as transporters (e.g., ABCB1) and tight junction proteins (e.g., ZO-1, claudin-5, and occludin) [124]. The presence of BBB is pivotal because the impairment of BBB is spotted in AD development, allowing the movement of immune cells and cytokines into the brain which is further followed by inflammatory events [128].

### 3.5.2 Parkinson's Disease

As in AD, several in vitro models have been designed and developed to study the pathophysiological aspects of PD and to test drug candidates. Some conventional 2D culture models used are HEK293, H4, PC12, and SH-SY5Y [33, 116]. Of those models, the latter is preferred as it displays a dopaminergic phenotype as seen in PD [129] and also mediates the formation of Lewy body-like inclusion after being exposed to human α-synuclein [130]. However, this cell line has a problem in reaching its postmitotic dopaminergic state [131]. As a result, SH-SY5Y can differentiate not only into dopaminergic neurons, but also other different neuronal cells [129].

The Lund human mesencephalic (LUHMES) cell line is another in vitro model taking more attention recently. These cells were isolated from healthy human fetal ventral mesencephalic tissue at 8 weeks old and were immortalized via *v-myc* insertion regulated by a tetracycline-responsive promoter [33, 116, 132]. LUHMES cells provide advantages in PD research as they can reach postmitotic state to differentiate into dopaminergic neurons similar to primary neurons [33, 116, 133]. The maturation of the neurons enriched with glial cells (e.g., astrocytes and oligodendrocytes) takes only 25 days [33, 134]. This cell line also possesses an ability to form an extracellular matrix naturally which is important in supporting cell interactions [133, 134].

The immortalized cell lines described above, like other cell lines, show a reduction in their properties along with the longer passage used. At this point, the use of primary culture isolated from human brains is an alternative. However, due to its

several limitations, the model is not always preferred. In addition to the ethical issues and the availability of the samples, the preparation and isolation of dopaminergic neurons from aged human brains are challenging. Looking at these drawbacks, dopaminergic neurons are mainly isolated from the embryonic brain of rodents [135]. However, the experiments utilizing primary culture may produce variable data depending on how the researchers dissect the brain out and prepare the isolation [33].

Furthermore, the use of iPSC and organoids for studying PD neuropathology and testing novel candidates for PD treatment is promising [33]. Although both models are directly derived from patients, making them close to the actual human cellular physiology, they also face limitations in their utilization, e.g., costly preparation, sophisticated procedures, and time-consuming [33]. However, the advantages of both models outweigh their disadvantages as described below.

It has been found that G2019S mutation occurring in leucine-rich repeat kinase 2 (LRRK2) plays a key role in the etiology of both familial and sporadic PD [136–140]. Upon these published findings, the concept of genetic role in late-onset PD changed. Previously, the genetic role in PD was only associated with several mutated genes, such as *PARKIN and PINK1*, which have been found frequently in early-onset PD, while the role of genetic mutation in sporadic PD was neglected [141]. This new milestone in studying PD becomes a trigger to design and develop a new PD model expressing LRRK2. As a result, both in vitro and in vivo models were successfully engineered so that they expressed mutated LRRK2. However, some LRRK2 mutation models fail to show dopaminergic neuron loss as well as Lewy body formation [142–144].

Furthermore, the human midbrain organoid derived from a patient suffering from sporadic PD is found to have LRRK2 G2019S mutation with dopaminergic neuronal loss [145, 146]. Another finding was reported by Kim and colleagues demonstrating that the length of dopaminergic neurites of isogenic midbrain organoids was reduced in the LRRK2 mutation group [143]. This was supported by the lower level of several important dopaminergic neuron markers, such as DAT, AADC, VMAT2, and TH, in the LRRK mutation group compared to the control group [143]. Further, this group also found that α-synuclein clearance was inhibited resulting in the aggregation of this protein inside the model [143].

The improvement of brain organoid models is underway to complement some shortcomings. As the role of glial cells and other supporting neuronal cells in PD pathophysiology is clear, then the model should cover this issue. Fortunately, Kwak et al. reported their success in generating midbrain organoid from human pluripotent stem cells that has the ability to differentiate into astrocytes, oligodendrocytes, and other supporting neuronal cells [147]. This group also confirmed that the model could produce midbrain dopaminergic neurons distributed homogenously while the exposure of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP), a parkinsonian neurotoxin [148], killed those dopaminergic neurons [147]. This confirms the functionality of the models for being an appropriate model system in studying PD pathophysiology or testing drug candidates.

Another issue that should be of concern is the role of the blood-brain barrier in PD progression. It has been known that BBB experiences increased leakage in PD as reflected by the higher value of the transfer coefficient across the BBB of a contrast substance in the PD group compared to the control group [149]. This could be caused by the impairment of ABC transporters, such as ABCB1, as shown by Kortekaas and coworkers who conducted an in vivo study in the parkinsonian midbrain [150]. Therefore, an in vitro model of PD should reflect the existence of the BBB. Cakir et al. have developed vascularized human brain organoids expressing key biomarkers of the BBB such as tight junction proteins (e.g., claudin-5, occludin, and ZO-1) and pivotal transporters (e.g., ABCB1 and GLUT1) [124]. Another group has also reported their success in generating vascularized brain organoids developed from iPSC-endothelial cells [151]. The existence of the vascular system in human brain organoids plays a critical role as the system could assist the development of the models by regulating neural differentiation and distribution, providing oxygen and nutrients, and helping in the formation of the neural circuit [127].

Although great success has been achieved recently in developing suitable in vitro models for PD, the use of animal models is still unavoidable. Some aspects that cannot be covered by in vitro model can be fulfilled by in vivo approach. For example, human physiological complexity can only be mimicked by animal models, especially mammals and nonhuman primates [115]. In this aspect, human organoids are also only partly suitable for covering the complexity of the human physiological system [115]. Nonhuman primates are a valuable model for studying PD pathology as aged primates, such as monkeys, show deficits in their nigrostriatal system and disturbances in α-synuclein distribution followed by the emergence of Parkinson-related symptoms [152, 153].

Numerous animal models have been developed and prepared as a platform for PD either as neurotoxin models, transgenic α-synuclein models, or models of other related genetic forms, such as LRRK2, of PD [103, 154]. Typically, the pharmacologic-based model of PD could be modeled by using two substances, namely, 6-hydroxydopamine (6-OHDA) and 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP). While the former substance does not readily cross the BBB, MPTP can cross the BBB as this substance is highly lipophilic. The uptake of both substances into the cell is mediated by the dopamine transporter system. Once inside the cell, they disturb the functions of mitochondria leading to excessive radical species production and ultimately cause neurodegeneration.

Several studies confirmed that the administration of MPTP to nonhuman primates produced parkinsonian symptoms such as bradykinesia, tremor, rigidity, cognitive deficits, and abnormalities in their posture and balance [155–157]. These symptoms were mainly related to the loss of dopaminergic neurons in the substantia nigra pars compacta area [155, 156], while Villalba et al. reported neuronal loss in the caudal intralaminar thalamic nuclei [157]. Nevertheless, the success in using nonhuman primates as a neurotoxin model is not always followed by the success in translating the achievements into the clinical setting [158], leaving researchers to keep the efforts for designing appropriate models that have good translatability.

One of the important requirements that must be fulfilled as a model of PD is the formation of aggregated α-synuclein. However, the formation of the aggregates gives inconsistent results in various models [154]. The aggregation of α-synuclein followed by its insertion into the Lewy body needs time. To overcome this time-consuming process, the use of the truncated c-terminal form of α-synuclein has been introduced. It has been found that the aggregation of α-synuclein in transgenic mice expressing the truncated proteins becomes faster than the wild-type group resulting in the progressive loss of dopaminergic neurons [154, 159, 160]. Interestingly, the truncation of the c-terminal of α-synuclein is found in a normal event in the human brain [161, 162].

### 3.5.3 Amyotrophic Lateral Sclerosis

Several genetic mutations have been identified as the major etiology in ALS generation and progression. Of those genes, mutations occurring in *tardbp* and *SOD1* genes have attracted higher interest as they are mostly found in either sporadic or familial ALS cases [86, 93]. Consequently, in the effort of designing ALS in vivo and in vitro models, these genes should be expressed by the chosen models. Furthermore, as ALS attacks motor neurons, any models used should exhibit the characteristic of this neuron [116].

Transgenic mice overexpressing SOD1 mutations have been developed to study the pathomechanism of ALS and to test the promising ALS drugs. Due to these mutations, the models experience significant motor neuron loss, excessive production and accumulation of misfolded SOD1, and ultimately paralysis occurring progressively [163, 164]. However, the inability of the SOD1 models in displaying the pathogenic aspect of TDP-43 has been reported and this becomes a shortcoming of these models [103].

In addition to mice, other animal models have generated interest. Nagai and colleagues reported the use of transgenic rats expressing human SOD1 that experienced progressive motor neuron degeneration [165]. Other animal models including marmoset [166], *Danio rerio* [167], *Drosophila melanogaster* [168], and *Caenorhabditis elegans* [169] have also been used in studying ALS.

As a DNA-/RNA-binding protein, TDP-43 is involved in numerous metabolic RNA processing targeting hundreds of molecules [170]. At this point, the development of pathological TDP-43 models is challenging as the targets of RNA processing mediated by TDP-43 are different between species [103]. Recently, approximately 20 transgenic rodent models of TDP-43 have been developed [171], while other transgenic animal models, such as nonhuman primates [172], zebrafish [167], fruit fly [173], and *Caenorhabditis elegans* [174], are also attracting attention.

Neurotoxin-based models of ALS have been developed. Recently, the use of bisphenol A exposed to zebrafish has been reported to induce degeneration of motor neurons even though this model has not been validated yet regarding the existence of ALS-related gene mutation such as *TDP-43* expressed by the model [175]. The

exposure of β-sitosterol-β-d-glucoside (BSSG) in murine has also been reported as one of the neurotoxin-based models [176].

Recently, the use of human organoids is becoming a powerful tool for studying ALS. Because ALS is characterized by progressive loss of motor neurons in the brain (upper motor neurons) and spinal cord (lower motor neurons), the development of organoid models should consider these sites. Pereira et al. confirmed their success in developing sensorimotor organoids generated from human iPSC lines isolated from ALS patients that can form neuromuscular junctions [177]. The human spinal cord organoid has also been developed [178, 179].

However, the promising use of organoid models in elucidating the fate of ALS should inspire researchers to improve their features from some drawbacks. For example, as the severity of ALS is strongly linked to the activation of glial cells, especially astrocytes and microglia [180], the development of an organoid model completed with glial cells is crucial. Vascularization of the organoids should also be considered. It has been reported that lack of a vascular system for a longer period of culture processes in organoids could lead to cell death as no sufficient oxygen and nutrients are supplied [127, 144]. Furthermore, BBB properties should be given more attention in the effort of improving brain organoid features as the impairment of the BBB is involved in ALS pathophysiology [181, 182]. Recently, Cakir and colleagues reported their achievement in generating brain organoids possessing the major features of the BBB such as ABC transporters (e.g., ABCB1 and GLUT1) and tight junctional proteins (e.g., occludin and claudin-5) [124]. This organoid model also expresses astrocytes and pericytes which are also critical in regulating barrier functions of the BBB [124].

## 3.6 Concluding Remarks and Future Directions

A close interrelationship between dysfunctional neurons and immune signaling pathways during the emergence and development of NDD has attracted much interest recently. This complex interplay brings several important consequences, including in the effort of developing novel drug candidates for NDDs that act on regulating immune cascades. Related to this consequence, immunotherapy use for treating NDDs has become an attractive idea nowadays. This approach basically utilizes the immune system to assist clearance of the pathogenic protein aggregates and removal of the noxious neurotoxins. Although most of the efforts are not successful, this insight may lead to more extensive studies in the future [183].

In addition, the failure in translating preclinical findings into clinical settings needs to be critically addressed. Most of the laboratory-based work uses both in vitro and in vivo models to represent the physiological and pathological aspects of NDDs. However, those models are not a complete representative to depict NDD in humans. Recently, the use of more physiologically relevant models, such as human organoid models and humanized chimeric models, is a promising progress. Further, the effort of developing microglia-like cells from iPSC has attracted much attention as these cells can depict the alteration of an immune signaling pathway associated with

NDDs. However, this model is not free from drawbacks. In general, the use of iPSC, including iPSC microglia, from different patients should consider the effect of genetic variation before going further to the conclusion [32]. It is noteworthy that a combination between the iPSC and other brain cell models, such as 3D brain organoids, may bring beneficial effects for deciphering the complex interaction among the cells which is not observable in 2D in vitro models [184, 185]. In principle, no model can entirely recapitulate both human physiology and pathophysiological aspects of NDDs in human [32]. Nonetheless, every effort conducted to reveal the NDD-related mysteries by using various models must be appreciated while at the same time the effort to develop a better model should also be carried on.

To sum up, there are still many unanswered questions regarding the role of inflammation in mediating the emergence and progression of NDDs. However, the interplay between NDDs and inflammation becomes evident. As a consequence, more resources should be allocated to investigate deeper about this topic of interest. An in-depth understanding gained regarding this topic would bring a brighter insight for tackling burdens caused by NDDs.

## References

1. Katsnelson A, De Strooper B, Zoghbi HY. Neurodegeneration: from cellular concepts to clinical applications. Sci Transl Med. 2016;8(364):364ps18.
2. Prince MJ, Wimo A, Guerchet MM, Ali GC, Wu Y-T, Prina M. World Alzheimer report 2015—the global impact of dementia: an analysis of prevalence, incidence, cost and trends; 2015.
3. United Nations DoE, Social Affairs PD. World population ageing 2015 report. ST/ESA/SER A/390; 2015.
4. Cummings JL, Morstorf T, Zhong K. Alzheimer's disease drug-development pipeline: few candidates, frequent failures. Alzheimers Res Ther. 2014;6(4):1–7.
5. Akhtar A, Andleeb A, Waris TS, Bazzar M, Moradi A-R, Awan NR, et al. Neurodegenerative diseases and effective drug delivery: a review of challenges and novel therapeutics. J Control Release. 2021;330:1152–67.
6. Cuny GD. Neurodegenerative diseases: challenges and opportunities. Future Med Chem. 2012;4(13):1647–9.
7. Rosemann A. Stem cell treatments for neurodegenerative diseases: challenges from a science, business and healthcare perspective. Neurodegener Dis manag. 2015;5(2):85–7.
8. Nainu F, Salim E, Asri RM, Hori A, Kuraishi T. Neurodegenerative disorders and sterile inflammation: lessons from a drosophila model. J Biochem. 2019;166(3):213–21.
9. Wyss-Coray T. Ageing, neurodegeneration and brain rejuvenation. Nature. 2016;539(7628):180–6.
10. Franceschi C, Garagnani P, Parini P, Giuliani C, Santoro A. Inflammaging: a new immune–metabolic viewpoint for age-related diseases. Nat Rev Endocrinol. 2018;14(10):576–90.
11. Xia S, Zhang X, Zheng S, Khanabdali R, Kalionis B, Wu J, et al. An update on inflamm-aging: mechanisms, prevention, and treatment. J Immunol Res. 2016;2016:8426874.
12. Walker LC, Jucker M. Neurodegenerative diseases: expanding the prion concept. Annu Rev Neurosci. 2015;38:87–103.
13. Sanders DW, Kaufman SK, DeVos SL, Sharma AM, Mirbaha H, Li A, et al. Distinct tau prion strains propagate in cells and mice and define different tauopathies. Neuron. 2014;82(6):1271–88.

14. Clavaguera F, Akatsu H, Fraser G, Crowther RA, Frank S, Hench J, et al. Brain homogenates from human tauopathies induce tau inclusions in mouse brain. Proc Natl Acad Sci. 2013;110 (23):9535–40.
15. Kaufman SK, Sanders DW, Thomas TL, Ruchinskas AJ, Vaquer-Alicea J, Sharma AM, et al. Tau prion strains dictate patterns of cell pathology, progression rate, and regional vulnerability in vivo. Neuron. 2016;92(4):796–812.
16. Iovino M, Agathou S, González-Rueda A, Del Castillo V-HM, Borroni B, Alberici A, et al. Early maturation and distinct tau pathology in induced pluripotent stem cell-derived neurons from patients with MAPT mutations. Brain. 2015;138(11):3345–59.
17. Honig LS, Vellas B, Woodward M, Boada M, Bullock R, Borrie M, et al. Trial of solanezumab for mild dementia due to Alzheimer's disease. N Engl J Med. 2018;378(4):321–30.
18. Murphy MP. Amyloid-beta solubility in the treatment of Alzheimer's disease. 2018
19. Richards RI, Robertson SA, Kastner DL. Neurodegenerative diseases have genetic hallmarks of autoinflammatory disease. Hum Mol Genet. 2018;27(R2):R108–R18.
20. Chitnis T, Weiner HL. CNS inflammation and neurodegeneration. J Clin Invest. 2017;127(10): 3577–87.
21. Fakhoury M. Immune-mediated processes in neurodegeneration: where do we stand? J Neurol. 2016;263(9):1683–701.
22. Bachiller S, Jiménez-Ferrer I, Paulus A, Yang Y, Swanberg M, Deierborg T, et al. Microglia in neurological diseases: a road map to brain-disease dependent-inflammatory response. Front Cell Neurosci. 2018;12:488.
23. Kwon HS, Koh SH. Neuroinflammation in neurodegenerative disorders: the roles of microglia and astrocytes. Transl Neurodegener. 2020;9(1):42.
24. Chen L, Deng H, Cui H, Fang J, Zuo Z, Deng J, et al. Inflammatory responses and inflammation-associated diseases in organs. Oncotarget. 2017;9(6):7204–18.
25. Glass CK, Saijo K, Winner B, Marchetto MC, Gage FH. Mechanisms underlying inflammation in neurodegeneration. Cell. 2010;140(6):918–34.
26. Amor S, Woodroofe MN. Innate and adaptive immune responses in neurodegeneration and repair. Immunology. 2014;141(3):287–91.
27. Griffin DE. Immune responses to RNA-virus infections of the CNS. Nat Rev Immunol. 2003;3 (6):493–502.
28. Klein RS, Garber C, Howard N. Infectious immunity in the central nervous system and brain function. Nat Immunol. 2017;18(2):132–41.
29. Zhang S-Y, Harschnitz O, Studer L, Casanova J-L. Neuron-intrinsic immunity to viruses in mice and humans. Curr Opin Immunol. 2021;72:309–17.
30. Sankowski R, Mader S, Valdés-Ferrer SI. Systemic inflammation and the brain: novel roles of genetic, molecular, and environmental cues as drivers of neurodegeneration. Front Cell Neurosci. 2015;9:28.
31. Stephenson J, Nutma E, van der Valk P, Amor S. Inflammation in CNS neurodegenerative diseases. Immunology. 2018;154(2):204–19.
32. Hammond TR, Marsh SE, Stevens B. Immune signaling in neurodegeneration. Immunity. 2019;50(4):955–74.
33. Slanzi A, Iannoto G, Rossi B, Zenaro E, Constantin G. In vitro models of neurodegenerative diseases. Front Cell Dev Biol. 2020;8:328.
34. Chaney A, Williams SR, Boutin H. In vivo molecular imaging of neuroinflammation in Alzheimer's disease. J Neurochem. 2019;149(4):438–51.
35. Ransohoff RM. How neuroinflammation contributes to neurodegeneration. Science. 2016;353 (6301):777–83.
36. Liddelow S, Barres B. SnapShot: astrocytes in health and disease. Cell. 2015;162(5):1170–e1.
37. Brettschneider J, Del Tredici K, Lee VM, Trojanowski JQ. Spreading of pathology in neurodegenerative diseases: a focus on human studies. Nat Rev Neurosci. 2015;16(2):109–20.

38. Wolfe MS. Chapter 1—solving the puzzle of neurodegeneration. In: Wolfe MS, editor. The molecular and cellular basis of neurodegenerative diseases. United State: Academic Press; 2018. p. 1–22.
39. Kovacs GG. Molecular pathology of neurodegenerative diseases: principles and practice. J Clin Pathol. 2019;72(11):725–35.
40. Dugger BN, Dickson DW. Pathology of neurodegenerative diseases. Cold Spring Harb Perspect Biol. 2017;9(7):a028035.
41. Takahashi RH, Nagao T, Gouras GK. Plaque formation and the intraneuronal accumulation of β-amyloid in Alzheimer's disease. Pathol Int. 2017;67(4):185–93.
42. Sun X, Chen WD, Wang YD. β-Amyloid: the key peptide in the pathogenesis of Alzheimer's disease. Front Pharmacol. 2015;6:221.
43. Terzi E, Hölzemann G, Seelig J. Interaction of Alzheimer beta-amyloid peptide(1-40) with lipid membranes. Biochemistry. 1997;36(48):14845–52.
44. Bishop GM, Robinson SR. Physiological roles of amyloid-beta and implications for its removal in Alzheimer's disease. Drugs Aging. 2004;21(10):621–30.
45. Hardy J. The amyloid hypothesis for Alzheimer's disease: a critical reappraisal. J Neurochem. 2009;110(4):1129–34.
46. Mota SI, Ferreira IL, Rego AC. Dysfunctional synapse in Alzheimer's disease—a focus on NMDA receptors. Neuropharmacology. 2014;76(Pt A):16–26.
47. Deane R, Du Yan S, Submamaryan RK, LaRue B, Jovanovic S, Hogg E, et al. RAGE mediates amyloid-beta peptide transport across the blood-brain barrier and accumulation in brain. Nat Med. 2003;9(7):907–13.
48. Donahue JE, Flaherty SL, Johanson CE, Duncan JA 3rd, Silverberg GD, Miller MC, et al. RAGE, LRP-1, and amyloid-beta protein in Alzheimer's disease. Acta Neuropathol. 2006;112 (4):405–15.
49. Kayed R, Head E, Thompson JL, McIntire TM, Milton SC, Cotman CW, et al. Common structure of soluble amyloid oligomers implies common mechanism of pathogenesis. Science. 2003;300(5618):486–9.
50. Zlokovic BV. The blood-brain barrier in health and chronic neurodegenerative disorders. Neuron. 2008;57(2):178–201.
51. Bertram L, Tanzi RE. Thirty years of Alzheimer's disease genetics: the implications of systematic meta-analyses. Nat Rev Neurosci. 2008;9(10):768–78.
52. Verghese PB, Castellano JM, Garai K, Wang Y, Jiang H, Shah A, et al. ApoE influences amyloid-β (Aβ) clearance despite minimal apoE/Aβ association in physiological conditions. Proc Natl Acad Sci U S A. 2013;110(19):E1807–16.
53. Akiyama H, Barger S, Barnum S, Bradt B, Bauer J, Cole GM, et al. Inflammation and Alzheimer's disease. Neurobiol Aging. 2000;21(3):383–421.
54. Kitazawa M, Yamasaki TR, LaFerla FM. Microglia as a potential bridge between the amyloid beta-peptide and tau. Ann N Y Acad Sci. 2004;1035:85–103.
55. Cartier L, Hartley O, Dubois-Dauphin M, Krause KH. Chemokine receptors in the central nervous system: role in brain inflammation and neurodegenerative diseases. Brain Res Brain Res Rev. 2005;48(1):16–42.
56. Griffin WS, Stanley LC, Ling C, White L, MacLeod V, Perrot LJ, et al. Brain interleukin 1 and S-100 immunoreactivity are elevated in down syndrome and Alzheimer disease. Proc Natl Acad Sci U S A. 1989;86(19):7611–5.
57. Landreth GE, Reed-Geaghan EG. Toll-like receptors in Alzheimer's disease. Curr Top Microbiol Immunol. 2009;336:137–53.
58. Reed-Geaghan EG, Savage JC, Hise AG, Landreth GE. CD14 and toll-like receptors 2 and 4 are required for fibrillar a{beta}-stimulated microglial activation. J Neurosci. 2009;29(38): 11982–92.
59. Schroder K, Tschopp J. The inflammasomes. Cell. 2010;140(6):821–32.
60. Yan SD, Chen X, Fu J, Chen M, Zhu H, Roher A, et al. RAGE and amyloid-β peptide neurotoxicity in Alzheimer's disease. Nature. 1996;382(6593):685–91.

61. Sastre M, Walter J, Gentleman SM. Interactions between APP secretases and inflammatory mediators. J Neuroinflammation. 2008;5:25.
62. Saijo K, Winner B, Carson CT, Collier JG, Boyer L, Rosenfeld MG, et al. A Nurr1/CoREST pathway in microglia and astrocytes protects dopaminergic neurons from inflammation-induced death. Cell. 2009;137(1):47–59.
63. McCoy MK, Tansey MG. TNF signaling inhibition in the CNS: implications for normal brain function and neurodegenerative disease. J Neuroinflammation. 2008;5:45.
64. Simi A, Tsakiri N, Wang P, Rothwell NJ. Interleukin-1 and inflammatory neurodegeneration. Biochem Soc Trans. 2007;35(Pt 5):1122–6.
65. Lister MF, Sharkey J, Sawatzky DA, Hodgkiss JP, Davidson DJ, Rossi AG, et al. The role of the purinergic P2X7 receptor in inflammation. J Inflamm (Lond). 2007;4:5.
66. Rissman RA, De Blas AL, Armstrong DM. GABA(a) receptors in aging and Alzheimer's disease. J Neurochem. 2007;103(4):1285–92.
67. Yamin G. NMDA receptor-dependent signaling pathways that underlie amyloid beta-protein disruption of LTP in the hippocampus. J Neurosci Res. 2009;87(8):1729–36.
68. Burré J, Sharma M, Südhof TC. Cell biology and pathophysiology of α-synuclein. Cold Spring Harb Perspect Med. 2018;8(3):a024091.
69. Mahul-Mellier AL, Burtscher J, Maharjan N, Weerens L, Croisier M, Kuttler F, et al. The process of Lewy body formation, rather than simply α-synuclein fibrillization, is one of the major drivers of neurodegeneration. Proc Natl Acad Sci U S A. 2020;117(9):4971–82.
70. Kim WS, Kågedal K, Halliday GM. Alpha-synuclein biology in Lewy body diseases. Alzheimers Res Ther. 2014;6(5):73.
71. Zhou J, Broe M, Huang Y, Anderson JP, Gai WP, Milward EA, et al. Changes in the solubility and phosphorylation of α-synuclein over the course of Parkinson's disease. Acta Neuropathol. 2011;121(6):695–704.
72. Lee JT, Wheeler TC, Li L, Chin LS. Ubiquitination of alpha-synuclein by Siah-1 promotes alpha-synuclein aggregation and apoptotic cell death. Hum Mol Genet. 2008;17(6):906–17.
73. Liu Y, Qiang M, Wei Y, He R. A novel molecular mechanism for nitrated {alpha}-synuclein-induced cell death. J Mol Cell Biol. 2011;3(4):239–49.
74. Nonaka T, Iwatsubo T, Hasegawa M. Ubiquitination of alpha-synuclein. Biochemistry. 2005;44(1):361–8.
75. Anderson JP, Walker DE, Goldstein JM, de Laat R, Banducci K, Caccavello RJ, et al. Phosphorylation of Ser-129 is the dominant pathological modification of alpha-synuclein in familial and sporadic Lewy body disease. J Biol Chem. 2006;281(40):29739–52.
76. Nakamura T, Yamashita H, Takahashi T, Nakamura S. Activated fyn phosphorylates alpha-synuclein at tyrosine residue 125. Biochem Biophys Res Commun. 2001;280(4):1085–92.
77. Okochi M, Walter J, Koyama A, Nakajo S, Baba M, Iwatsubo T, et al. Constitutive phosphorylation of the Parkinson's disease associated alpha-synuclein. J Biol Chem. 2000;275(1):390–7.
78. Roodveldt C, Christodoulou J, Dobson CM. Immunological features of alpha-synuclein in Parkinson's disease. J Cell Mol Med. 2008;12(5b):1820–9.
79. Wolozin B, Behl C. Mechanisms of neurodegenerative disorders: part 1: protein aggregates. Arch Neurol. 2000;57(6):793–6.
80. Lashuel HA, Overk CR, Oueslati A, Masliah E. The many faces of α-synuclein: from structure and toxicity to therapeutic target. Nat Rev Neurosci. 2013;14(1):38–48.
81. McGeer PL, McGeer EG. Glial reactions in Parkinson's disease. Mov Disord. 2008;23(4):474–83.
82. Hirsch EC, Hunot S. Neuroinflammation in Parkinson's disease: a target for neuroprotection? Lancet Neurol. 2009;8(4):382–97.
83. Kim WG, Mohney RP, Wilson B, Jeohn GH, Liu B, Hong JS. Regional difference in susceptibility to lipopolysaccharide-induced neurotoxicity in the rat brain: role of microglia. J Neurosci. 2000;20(16):6309–16.

84. Dong Y, Chen Y. The role of ubiquitinated TDP-43 in amyotrophic lateral sclerosis. Neuroimmunology and Neuroinflammation. 2018;5:5.
85. Arai T, Hasegawa M, Akiyama H, Ikeda K, Nonaka T, Mori H, et al. TDP-43 is a component of ubiquitin-positive tau-negative inclusions in frontotemporal lobar degeneration and amyotrophic lateral sclerosis. Biochem Biophys Res Commun. 2006;351(3):602–11.
86. Kabashi E, Valdmanis PN, Dion P, Spiegelman D, McConkey BJ, Vande Velde C, et al. TARDBP mutations in individuals with sporadic and familial amyotrophic lateral sclerosis. Nat Genet. 2008;40(5):572–4.
87. Lim L, Wei Y, Lu Y, Song J. ALS-causing mutations significantly perturb the self-assembly and interaction with nucleic acid of the intrinsically disordered prion-like domain of TDP-43. PLoS Biol. 2016;14(1):e1002338.
88. Neumann M, Sampathu DM, Kwong LK, Truax AC, Micsenyi MC, Chou TT, et al. Ubiquitinated TDP-43 in frontotemporal lobar degeneration and amyotrophic lateral sclerosis. Science. 2006;314(5796):130–3.
89. Ayala YM, Zago P, D'Ambrogio A, Xu YF, Petrucelli L, Buratti E, et al. Structural determinants of the cellular localization and shuttling of TDP-43. J Cell Sci. 2008;121 (Pt 22):3778–85.
90. Barmada SJ, Skibinski G, Korb E, Rao EJ, Wu JY, Finkbeiner S. Cytoplasmic mislocalization of TDP-43 is toxic to neurons and enhanced by a mutation associated with familial amyotrophic lateral sclerosis. J Neurosci. 2010;30(2):639–49.
91. Barmada SJ, Serio A, Arjun A, Bilican B, Daub A, Ando DM, et al. Autophagy induction enhances TDP43 turnover and survival in neuronal ALS models. Nat Chem Biol. 2014;10(8): 677–85.
92. Iguchi Y, Eid L, Parent M, Soucy G, Bareil C, Riku Y, et al. Exosome secretion is a key pathway for clearance of pathological TDP-43. Brain. 2016;139(Pt 12):3187–201.
93. Rosen DR, Siddique T, Patterson D, Figlewicz DA, Sapp P, Hentati A, et al. Mutations in cu/Zn superoxide dismutase gene are associated with familial amyotrophic lateral sclerosis. Nature. 1993;362(6415):59–62.
94. Siddique T, Figlewicz DA, Pericak-Vance MA, Haines JL, Rouleau G, Jeffers AJ, et al. Linkage of a gene causing familial amyotrophic lateral sclerosis to chromosome 21 and evidence of genetic-locus heterogeneity. N Engl J Med. 1991;324(20):1381–4.
95. Kwiatkowski TJ Jr, Bosco DA, Leclerc AL, Tamrazian E, Vanderburg CR, Russ C, et al. Mutations in the FUS/TLS gene on chromosome 16 cause familial amyotrophic lateral sclerosis. Science. 2009;323(5918):1205–8.
96. Vance C, Rogelj B, Hortobágyi T, De Vos KJ, Nishimura AL, Sreedharan J, et al. Mutations in FUS, an RNA processing protein, cause familial amyotrophic lateral sclerosis type 6. Science. 2009;323(5918):1208–11.
97. Huang WJ, Chen WW, Zhang X. Multiple sclerosis: pathology, diagnosis and treatments. Exp Ther Med. 2017;13(6):3163–6.
98. Castro G, Liu X, Ngo K, De Leon-Tabaldo A, Zhao S, Luna-Roman R, et al. RORγt and RORα signature genes in human Th17 cells. PLoS One. 2017;12(8):e0181868.
99. Ruan Q, Kameswaran V, Zhang Y, Zheng S, Sun J, Wang J, et al. The Th17 immune response is controlled by the Rel-RORγ-RORγ T transcriptional axis. J Exp Med. 2011;208(11): 2321–33.
100. Wen SR, Liu GJ, Feng RN, Gong FC, Zhong H, Duan SR, et al. Increased levels of IL-23 and osteopontin in serum and cerebrospinal fluid of multiple sclerosis patients. J Neuroimmunol. 2012;244(1–2):94–6.
101. Yu C, Kam W, Dambuza I, Marrero B, Mahdi R, Egwuagu CE, et al. Osteopontin is expressed by microglia and T cells and regulated by STAT3. Invest Ophthalmol Vis Sci. 2013;54(15): 2050.
102. Krumbholz M, Theil D, Derfuss T, Rosenwald A, Schrader F, Monoranu CM, et al. BAFF is produced by astrocytes and up-regulated in multiple sclerosis lesions and primary central nervous system lymphoma. J Exp Med. 2005;201(2):195–200.

103. Dawson TM, Golde TE, Lagier-Tourenne C. Animal models of neurodegenerative diseases. Nat Neurosci. 2018;21(10):1370–9.
104. Ashe KH, Zahs KR. Probing the biology of Alzheimer's disease in mice. Neuron. 2010;66(5): 631–45.
105. LaFerla FM, Green KN. Animal models of Alzheimer disease. Cold Spring Harb Perspect Med. 2012;2(11):a006320.
106. Bekris LM, Yu CE, Bird TD, Tsuang DW. Genetics of Alzheimer disease. J Geriatr Psychiatry Neurol. 2010;23(4):213–27.
107. Ribeiro FM, Camargos ER, de Souza LC, Teixeira AL. Animal models of neurodegenerative diseases. Braz J Psychiatry. 2013;35(Suppl 2):S82–91.
108. Reardon S. Frustrated Alzheimer's researchers seek better lab mice. Nature. 2018;563(7733): 611–2.
109. Li HW, Zhang L, Qin C. Current state of research on non-human primate models of Alzheimer's disease. Animal Model Exp Med. 2019;2(4):227–38.
110. Emborg ME. Nonhuman primate models of neurodegenerative disorders. ILAR J. 2017;58(2): 190–201.
111. Palazzi X, Switzer R, George C. Natural occurrence of amyloid-abeta deposits in the brain of young common marmosets (Callithrix jacchus): a morphological and immunohistochemical evaluation. Vet Pathol. 2006;43(5):777–9.
112. Bilen J, Bonini NM. Drosophila as a model for human neurodegenerative disease. Annu Rev Genet. 2005;39:153–71.
113. Laird AS, Robberecht W. Modeling neurodegenerative diseases in zebrafish embryos. Methods Mol Biol. 2011;793:167–84.
114. Li J, Le W. Modeling neurodegenerative diseases in Caenorhabditis elegans. Exp Neurol. 2013;250:94–103.
115. Kim J, Koo B-K, Knoblich JA. Human organoids: model systems for human biology and medicine. Nat Rev Mol Cell Biol. 2020;21(10):571–84.
116. Schlachetzki JC, Saliba SW, Oliveira AC. Studying neurodegenerative diseases in culture models. Braz J Psychiatry. 2013;35(Suppl 2):S92–100.
117. Israel MA, Yuan SH, Bardy C, Reyna SM, Mu Y, Herrera C, et al. Probing sporadic and familial Alzheimer's disease using induced pluripotent stem cells. Nature. 2012;482(7384): 216–20.
118. Li T, Pires C, Nielsen TT, Waldemar G, Hjermind LE, Nielsen JE, et al. Generation of induced pluripotent stem cells (iPSCs) from an Alzheimer's disease patient carrying an A79V mutation in PSEN1. Stem Cell Res. 2016;16(2):229–32.
119. Yagi T, Ito D, Okada Y, Akamatsu W, Nihei Y, Yoshizaki T, et al. Modeling familial Alzheimer's disease with induced pluripotent stem cells. Hum Mol Genet. 2011;20(23): 4530–9.
120. D'Avanzo C, Aronson J, Kim YH, Choi SH, Tanzi RE, Kim DY. Alzheimer's in 3D culture: challenges and perspectives. Bioessays. 2015;37(10):1139–48.
121. González-Reyes RE, Nava-Mesa MO, Vargas-Sánchez K, Ariza-Salamanca D, Mora-Muñoz L. Involvement of astrocytes in Alzheimer's disease from a neuroinflammatory and oxidative stress perspective. Front Mol Neurosci. 2017;10:427.
122. Nagele RG, Wegiel J, Venkataraman V, Imaki H, Wang KC, Wegiel J. Contribution of glial cells to the development of amyloid plaques in Alzheimer's disease. Neurobiol Aging. 2004;25(5):663–74.
123. Choi SH, Kim YH, Hebisch M, Sliwinski C, Lee S, D'Avanzo C, et al. A three-dimensional human neural cell culture model of Alzheimer's disease. Nature. 2014;515(7526):274–8.
124. Cakir B, Xiang Y, Tanaka Y, Kural MH, Parent M, Kang Y-J, et al. Engineering of human brain organoids with a functional vascular-like system. Nat Methods. 2019;16(11):1169–75.
125. Kelava I, Lancaster MA. Stem cell models of human brain development. Cell Stem Cell. 2016;18(6):736–48.

126. Shi Y, Sun L, Wang M, Liu J, Zhong S, Li R, et al. Vascularized human cortical organoids (vOrganoids) model cortical development in vivo. PLoS Biol. 2020;18(5):e3000705.
127. Matsui TK, Tsuru Y, Hasegawa K, Kuwako K-i. Vascularization of human brain organoids. Stem Cells. 2021;39(8):1017–24.
128. Cai Z, Qiao PF, Wan CQ, Cai M, Zhou NK, Li Q. Role of blood-brain barrier in Alzheimer's disease. J Alzheimers Dis. 2018;63(4):1223–34.
129. Xicoy H, Wieringa B, Martens GJM. The SH-SY5Y cell line in Parkinson's disease research: a systematic review. Mol Neurodegener. 2017;12(1):10.
130. Taylor-Whiteley TR, Le Maitre CL, Duce JA, Dalton CF, Smith DP. Recapitulating Parkinson's disease pathology in a three-dimensional human neural cell culture model. Dis Model Mech. 2019;12(4):dmm038042.
131. Constantinescu R, Constantinescu AT, Reichmann H, Janetzky B. Neuronal differentiation and long-term culture of the human neuroblastoma line SH-SY5Y. J Neural Transm Suppl. 2007;72:17–28.
132. Zhang XM, Yin M, Zhang MH. Cell-based assays for Parkinson's disease using differentiated human LUHMES cells. Acta Pharmacol Sin. 2014;35(7):945–56.
133. Scholz D, Pöltl D, Genewsky A, Weng M, Waldmann T, Schildknecht S, et al. Rapid, complete and large-scale generation of post-mitotic neurons from the human LUHMES cell line. J Neurochem. 2011;119(5):957–71.
134. Honegger P. Overview of cell and tissue culture techniques. Curr Protoc Pharmacol. 2001;-Chapter 12:Unit12.1.
135. Gaven F, Marin P, Claeysen S. Primary culture of mouse dopaminergic neurons. J Vis Exp. 2014;91:e51751.
136. Lesage S, Dürr A, Tazir M, Lohmann E, Leutenegger A-L, Janin S, et al. LRRK2 G2019S as a cause of Parkinson's disease in North african arabs. N Engl J Med. 2006;354(4):422–3.
137. Ozelius LJ, Senthil G, Saunders-Pullman R, Ohmann E, Deligtisch A, Tagliati M, et al. LRRK2 G2019S as a cause of Parkinson's disease in Ashkenazi Jews. N Engl J Med. 2006;354(4):424–5.
138. Gilks WP, Abou-Sleiman PM, Gandhi S, Jain S, Singleton A, Lees AJ, et al. A common LRRK2 mutation in idiopathic Parkinson's disease. Lancet. 2005;365(9457):415–6.
139. Goldwurm S, Di Fonzo A, Simons EJ, Rohé CF, Zini M, Canesi M, et al. The G6055A (G2019S) mutation in LRRK2 is frequent in both early and late onset Parkinson's disease and originates from a common ancestor. J Med Genet. 2005;42(11):e65.
140. Ferreira JJ, Guedes LC, Rosa MM, Coelho M, van Doeselaar M, Schweiger D, et al. High prevalence of LRRK2 mutations in familial and sporadic Parkinson's disease in Portugal. Mov Disord. 2007;22(8):1194–201.
141. Bonifati V. Parkinson's disease: the LRRK2-G2019S mutation: opening a novel era in Parkinson's disease genetics. Eur J Hum Genet. 2006;14(10):1061–2.
142. Chesselet MF, Fleming S, Mortazavi F, Meurers B. Strengths and limitations of genetic mouse models of Parkinson's disease. Parkinsonism Relat Disord. 2008;14 Suppl 2(Suppl 2):S84–7.
143. Kim H, Park HJ, Choi H, Chang Y, Park H, Shin J, et al. Modeling G2019S-LRRK2 sporadic Parkinson's disease in 3D midbrain organoids. Stem Cell Reports. 2019;12(3):518–31.
144. Shou Y, Liang F, Xu S, Li X. The application of brain organoids: from neuronal development to neurological diseases. Front Cell Dev Biol. 2020;8:579659.
145. Kordower JH, Olanow CW, Dodiya HB, Chu Y, Beach TG, Adler CH, et al. Disease duration and the integrity of the nigrostriatal system in Parkinson's disease. Brain. 2013;136(Pt 8): 2419–31.
146. Smits LM, Reinhardt L, Reinhardt P, Glatza M, Monzel AS, Stanslowsky N, et al. Modeling Parkinson's disease in midbrain-like organoids. Npj Parkinson's Disease. 2019;5(1):5.
147. Kwak TH, Kang JH, Hali S, Kim J, Kim KP, Park C, et al. Generation of homogeneous midbrain organoids with in vivo-like cellular composition facilitates neurotoxin-based Parkinson's disease modeling. Stem Cells. 2020;38(6):727–40.

148. Hazell AS, Itzhak Y, Liu H, Norenberg MD. 1-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) decreases glutamate uptake in cultured astrocytes. J Neurochem. 1997;68(5):2216–9.
149. Al-Bachari S, Naish JH, Parker GJM, Emsley HCA, Parkes LM. Blood–brain barrier leakage is increased in Parkinson's disease. Front Physiol. 2020;11(1636):593026.
150. Kortekaas R, Leenders KL, van Oostrom JC, Vaalburg W, Bart J, Willemsen AT, et al. Blood-brain barrier dysfunction in parkinsonian midbrain in vivo. Ann Neurol. 2005;57(2):176–9.
151. Pham MT, Pollock KM, Rose MD, Cary WA, Stewart HR, Zhou P, et al. Generation of human vascularized brain organoids. Neuroreport. 2018;29(7):588–93.
152. Emborg ME, Ma SY, Mufson EJ, Levey AI, Taylor MD, Brown WD, et al. Age-related declines in nigral neuronal function correlate with motor impairments in rhesus monkeys. J Comp Neurol. 1998;401(2):253–65.
153. Chu Y, Kordower JH. Age-associated increases of alpha-synuclein in monkeys and humans are associated with nigrostriatal dopamine depletion: is this the target for Parkinson's disease? Neurobiol Dis. 2007;25(1):134–49.
154. Bezard E, Yue Z, Kirik D, Spillantini MG. Animal models of Parkinson's disease: limits and relevance to neuroprotection studies. Mov Disord. 2013;28(1):61–70.
155. Burns RS, Chiueh CC, Markey SP, Ebert MH, Jacobowitz DM, Kopin IJ. A primate model of parkinsonism: selective destruction of dopaminergic neurons in the pars compacta of the substantia nigra by N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine. Proc Natl Acad Sci U S A. 1983;80(14):4546–50.
156. Langston JW, Forno LS, Rebert CS, Irwin I. Selective nigral toxicity after systemic administration of 1-methyl-4-phenyl-1,2,5,6-tetrahydropyrine (MPTP) in the squirrel monkey. Brain Res. 1984;292(2):390–4.
157. Villalba RM, Wichmann T, Smith Y. Neuronal loss in the caudal intralaminar thalamic nuclei in a primate model of Parkinson's disease. Brain Struct Funct. 2014;219(1):381–94.
158. Athauda D, Foltynie T. Challenges in detecting disease modification in Parkinson's disease clinical trials. Parkinsonism Relat Disord. 2016;32:1–11.
159. Tofaris GK, Garcia Reitböck P, Humby T, Lambourne SL, O'Connell M, Ghetti B, et al. Pathological changes in dopaminergic nerve cells of the substantia nigra and olfactory bulb in mice transgenic for truncated human alpha-synuclein(1-120): implications for Lewy body disorders. J Neurosci. 2006;26(15):3942–50.
160. Liu CW, Giasson BI, Lewis KA, Lee VM, Demartino GN, Thomas PJ. A precipitating role for truncated alpha-synuclein and the proteasome in alpha-synuclein aggregation: implications for pathogenesis of Parkinson disease. J Biol Chem. 2005;280(24):22670–8.
161. Li W, West N, Colla E, Pletnikova O, Troncoso JC, Marsh L, et al. Aggregation promoting C-terminal truncation of alpha-synuclein is a normal cellular process and is enhanced by the familial Parkinson's disease-linked mutations. Proc Natl Acad Sci U S A. 2005;102(6):2162–7.
162. Muntané G, Ferrer I, Martinez-Vicente M. α-Synuclein phosphorylation and truncation are normal events in the adult human brain. Neuroscience. 2012;200:106–19.
163. Gurney ME, Pu H, Chiu AY, Dal Canto MC, Polchow CY, Alexander DD, et al. Motor neuron degeneration in mice that express a human cu, Zn superoxide dismutase mutation. Science. 1994;264(5166):1772–5.
164. Philips T, Rothstein JD. Rodent models of amyotrophic lateral sclerosis. Curr Protoc Pharmacol. 2015;69:5.67.1–5.67.21.
165. Nagai M, Aoki M, Miyoshi I, Kato M, Pasinelli P, Kasai N, et al. Rats expressing human cytosolic copper-zinc superoxide dismutase transgenes with amyotrophic lateral sclerosis: associated mutations develop motor neuron disease. J Neurosci. 2001;21(23):9246–54.
166. Borel F, Gernoux G, Cardozo B, Metterville JP, Toro Cabrera GC, Song L, et al. Therapeutic rAAVrh10 mediated SOD1 silencing in adult SOD1(G93A) mice and nonhuman primates. Hum Gene Ther. 2016;27(1):19–31.
167. Morrice JR, Gregory-Evans CY, Shaw CA. Animal models of amyotrophic lateral sclerosis: a comparison of model validity. Neural Regen Res. 2018;13(12):2050–4.

168. Watson MR, Lagow RD, Xu K, Zhang B, Bonini NM. A drosophila model for amyotrophic lateral sclerosis reveals motor neuron damage by human SOD1. J Biol Chem. 2008;283(36): 24972–81.
169. Cornaglia M, Krishnamani G, Mouchiroud L, Sorrentino V, Lehnert T, Auwerx J, et al. Automated longitudinal monitoring of in vivo protein aggregation in neurodegenerative disease C. elegans models. Mol Neurodegener. 2016;11:17.
170. Prasad A, Bharathi V, Sivalingam V, Girdhar A, Patel BK. Molecular mechanisms of TDP-43 misfolding and pathology in amyotrophic lateral sclerosis. Front Mol Neurosci. 2019;12:25.
171. Lutz C. Mouse models of ALS: past, present and future. Brain Res. 2018;1693(Pt A):1–10.
172. Uchida A, Sasaguri H, Kimura N, Tajiri M, Ohkubo T, Ono F, et al. Non-human primate model of amyotrophic lateral sclerosis with cytoplasmic mislocalization of TDP-43. Brain. 2012;135(Pt 3):833–46.
173. Chang JC, Hazelett DJ, Stewart JA, Morton DB. Motor neuron expression of the voltage-gated calcium channel cacophony restores locomotion defects in a drosophila, TDP-43 loss of function model of ALS. Brain Res. 2014;1584:39–51.
174. Vaccaro A, Tauffenberger A, Aggad D, Rouleau G, Drapeau P, Parker JA. Mutant TDP-43 and FUS cause age-dependent paralysis and neurodegeneration in C. elegans. PLoS One. 2012;7(2):e31321.
175. Morrice JR, Gregory-Evans CY, Shaw CA. Modeling environmentally-induced motor neuron degeneration in zebrafish. Sci Rep. 2018;8(1):4890.
176. Wilson JM, Khabazian I, Wong MC, Seyedalikhani A, Bains JS, Pasqualotto BA, et al. Behavioral and neurological correlates of ALS-parkinsonism dementia complex in adult mice fed washed cycad flour. Neuromolecular Med. 2002;1(3):207–21.
177. Pereira JD, DuBreuil DM, Devlin A-C, Held A, Sapir Y, Berezovski E, et al. Human sensorimotor organoids derived from healthy and amyotrophic lateral sclerosis stem cells form neuromuscular junctions. Nat Commun. 2021;12(1):4744.
178. Hor JH, Soh ES-Y, Tan LY, Lim VJW, Santosa MM, Winanto, et al. Cell cycle inhibitors protect motor neurons in an organoid model of spinal muscular atrophy. Cell Death Dis. 2018;9(11):1100.
179. Winanto KZ-J, Hor J-H, Ng S-Y. Spinal cord organoids add an extra dimension to traditional motor neuron cultures. Neural Regen Res. 2019;14(9):1515–6.
180. Lasiene J, Yamanaka K. Glial cells in amyotrophic lateral sclerosis. Neurol Res Int. 2011;2011:718987.
181. Garbuzova-Davis S, Sanberg P. Blood-CNS barrier impairment in ALS patients versus an animal model. Front Cell Neurosci. 2014;8:21.
182. Kakaroubas N, Brennan S, Keon M, Saksena NK. Pathomechanisms of blood-brain barrier disruption in ALS. Neurosci J. 2019;2019:2537698.
183. Liu E, Schmidt ME, Margolin R, Sperling R, Koeppe R, Mason NS, et al. Amyloid-β 11C-PiB-PET imaging results from 2 randomized bapineuzumab phase 3 AD trials. Neurology. 2015;85(8):692–700.
184. Abud EM, Ramirez RN, Martinez ES, Healy LM, Nguyen CH, Newman SA, et al. iPSC-derived human microglia-like cells to study neurological diseases. Neuron. 2017;94(2): 278–93. e9.
185. Mancuso R, Van Den Daele J, Fattorelli N, Wolfs L, Balusu S, Burton O, et al. Stem-cell-derived human microglia transplanted in mouse brain to study human disease. Nat Neurosci. 2019;22(12):2111–6.

# Microbiome-Induced Autoimmunity and Novel Therapeutic Intervention

4

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**Abstract**

Microorganisms' flora, which colonize in many parts of our body, stand out as one of the most important components for a healthy life. This microbial organization called microbiome lives in integration with the body as a single and whole organ/system. Perhaps, the human first encounters the microbial activity it carries through the immune system. This encounter and interaction are vital for the development of immune system cells that protect the body against pathogenic organisms and infections throughout life. In recent years, it has been determined that some disruptions in the host-microbiome interaction play an important role in the physiopathology of autoimmune diseases. Although the details of this interaction have not been clarified yet, the focus is on leaky gut syndrome, dysbiosis, toll-like receptor ligands, and B cell dysfunction. Nutritional regulations, prebiotics, probiotics, fecal microbiota transplantation, bacterial engineering, and vaccination are being investigated as new therapeutic approaches in the treatment of problems in these areas. This article reviews recent research in this area.

**Keywords**

Microbiome · Autoimmunity · Neuroinflammation · Dysbiosis · Vaccination · Leaky gut

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## 4.1 Introduction

Studies examining the effect of the microbiome on health and the formation of diseases have yielded very important data on a strong and bidirectional interaction between bacteria and eukaryotic functions. This interaction, which is probably established in the intrauterine period and lasts for life, is not pathogenic but largely symbiotic.

Although the pathogenic effect of microorganisms was first revealed by Robert Koch in 1876, the first-century BC Roman scientist Marcus Terentius Varro predicted that microbes cause infections [1, 2]. In the following period, microbes were recognized only for their disease-causing roles. However, the first scientist to offer a different perspective on the subject was Nobel Prize winner Elie Metchnikoff. According to his hypothesis, there is a relationship between the beneficial microbes in yogurt and the strengthening of the immune system [3]. During the same years, J. George Porter Phillips drew attention to the potential of lactobacilli in the treatment of depression [4]. Although the importance of the subject was understood in those years, no further development could be put forward due to technological impossibilities.

With the "hygiene hypothesis," in 1989, David P. Strachan argued that there may be a correlation between antimicrobial applications and the increase in allergic diseases [5]. Then, Graham A. Rook developed the hygiene hypothesis and brought a broader interpretation to the relationship between unicellular and multicellular life. According to the "old friends hypothesis" proposed by Rook, human evolution is modulated and even stimulated by commensal microorganisms [6]. This is called "co-evolution."

Where on the host did these microorganisms colonize? In fact, a very large part of the host body is colonized by bacteria. They are present on the skin, starting from the mouth throughout the gastrointestinal tract, and in the vagina. The organ with the highest number of bacterial colonization is the intestines. It is estimated that approximately four times the number of human eukaryotic cells (380 trillion) prokaryotic microorganisms live in the intestines [7]. Organic materials belonging to these microorganisms are distributed in the intestinal lumen and these gene particles are mixed into the systemic circulation. Thus, prokaryotic materials come into contact with eukaryotic cells and processes coevolution [8–10].

## 4.2 Microbiome-Induced Autoimmunity

### 4.2.1 Microbiome

Prokaryotes are the first living cells to come to life on earth and they constitute the largest group among living things in terms of population [11]. *Homo sapiens* hosts millions of prokaryotic cells in addition to its own eukaryotic cells that carry 46 chromosomes [12]. There is hardly a region where microorganisms do not colonize the host body [13]. Our guests communicate with each other [14] and

with the eukaryotic cells of the host [15]. Although speculative, there are studies that talk about "brain microbiota" [16].

Commensal prokaryotes are most commonly colonized in the gastrointestinal tract, especially the colon, in the human body [17]. The total number of these microbiota prokaryotes is greater than the number of eukaryotic cells, and the number of genes they have is more than the number of human genes [10].

Microbiota composition may vary depending on the genetic structure, aging, and environmental factors (nutrition, stress level, medications). For example, olanzapine exposure increases Firmicutes levels and decreases Proteobacteria levels [18]. *Bifidobacteria* and *Lactobacillus* levels decrease after acute stress in newborns [19, 20]. Depending on chronic stress, *Bacteroides* decrease and *Clostridium* increases [21]. The long-term use of broad-spectrum antibiotics can permanently alter the composition of the gut microbiota [22]. These reasons make the microbiota composition dynamic and unique throughout life [23–25].

The immune system and microbes interact in a complex way. Prokaryotic cell wall elements and genetic materials stimulate immune system cells [17]. This interaction is mediated by pattern recognition receptors (PRRs) and toll-like receptors (TLRs) [26]. Commensal bacteria such as *Lactobacillus* GG and *Bifidobacterium infantis* create an anti-inflammatory effect by increasing the level of interleukin-10 [27, 28]. They can also inhibit the pro-inflammatory process by stimulating TLR-2 and TLR-4 [29].

### 4.2.2 Maternal Microbiota

During pregnancy, very interesting adaptations occur in the female body, perhaps the most important of which is the development of the placenta. One of the functions of the placenta is to prevent maternal immunogenicity towards the fetus (as well as the fetus towards the mother). This complex and highly specialized organ provides fetomaternal exchange of molecules. This exchange includes materials originating from the maternal microbiota [30].

In recent years, there has been a great deal of research and speculation that draws attention to the existence of a placental microbiota. Whether the placenta harbors a microbial community is still a matter of argument. These discussions started with a study conducted in 2014 [31]. In this study challenging the "sterile uterus" paradigm, human placenta samples were examined and a microbial colonization was detected [31]. In previous publications, bacteria were found in the human placenta during term [32, 33] and preterm [33] births. Studies that isolated bacteria from umbilical cord blood [34], meconium [35], and amniotic fluid [32] have also been published. In addition, it was isolated from culture of amniotic fluid [34] and meconium [35] following oral administration of genetically labeled *E. faecium* to pregnant mice. In the following period, many studies [36–39] claiming the existence of placental microbiota were conducted. Shortly after the article published by Aagaard et al. in 2014 [31], Kliman et al. claimed that the detection of bacterial DNA alone does not provide evidence for the presence of living microbes [40]. Over time, it became clear

that the contamination issues [41, 42] and the microbiome of the test kit (for which the name "kitome" was proposed) [43] posed great challenges in the search for a microbiota that lives in the placenta.

Later, further studies were conducted with more rigorous scrutiny at each step of the process (including only cesarean section tissue samples to reduce the risk of contamination, comparing bacterial taxa with those in the immediate environment, and removing taxa that overlap with the kitome) [44–48]. Despite all these measures, a unique placental microbiota could not be detected [41, 43, 49–52]. However, although there are recently published articles claiming to detect bacterial DNA and live bacteria in the fetal gut, this information still seems speculative [53–55].

With or without the placental microbiome, the effect of microorganisms on the immune system begins in the intrauterine period [56]. Toll-like receptors (TLRs) on intestinal epithelial cells are thought to be the first step in cytokine production [57, 58]. Intestinal microorganisms can directly affect cytokine production by contacting TLRs [59–61]. As a result of the stimulation of TLRs, the production of pro-inflammatory cytokines increases [62]. Dendritic cells, one of the cells of the immune system in the intestines, can take the bacteria in the intestinal lumen and their metabolites into the cytoplasm, causing them to mix into the systemic circulation and initiate an immune reaction [63]. The results of changes in the composition of the microbiota in the postnatal period can continue throughout life [64].

### 4.2.3 Hygiene Hypothesis

Strachan's original report in 1989 was based on a simple observation: hay fever and atopic dermatitis occur less frequently in families with many children than in families with only one or two children [5]. He suggested that there may be a negative correlation between the increase in the frequency of allergic diseases observed in the previous three or four decades and the decrease in the frequency of infectious diseases in the following period [65]. In the early 2000s, the hygiene hypothesis was expanded to include autoimmune diseases [66]. At the time, data were already available from experimental models showing that infections, particularly parasitic infections, could prevent the occurrence of autoimmunity [67]. To date, convincing evidence was gathered to support the hygiene hypothesis.

As with every hypothesis, there are opinions against the hygiene hypothesis. When the literature is searched, it is seen that the majority of several hundred articles on the subject support the hypothesis. However, there are still articles that question the hypothesis and express hesitations. It is questioned whether the recommendations of 20 years ago are still valid today [66].

It is clear that the incidence of communicable diseases is decreasing in industrialized countries where hygiene, vaccines, and antibiotics are widely used. However, new pandemics such as COVID-19 may occur. In addition, the frequency of type 1 diabetes mellitus and allergic diseases continues to increase in recent years [68–70].

Evidence of worsening asthma severity has been reported in patients exposed to antiparasitic treatments [71]. Although data are contradictory, we can talk about a decrease in atopic susceptibility after probiotic administration [72].

There are relatively few publications for autoimmune diseases. The first convincing observation that autoimmune diseases could be prevented by infestation of the parasite *Plasmodium berghei* was published in 1970 [67]. Recently, there are studies suggesting that *Trichuris suis* infestation has a positive effect on patients with multiple sclerosis [73, 74]. It has been shown that autoimmune diseases such as systemic lupus erythematosus can be significantly cured by administering viral [75] and parasitic [76] microorganisms in mouse experiments. Other studies have confirmed this positive effect of parasites [77–81].

However, the observation that there is a negative relationship between the decrease in infections and the increase in allergic and autoimmune diseases is not enough to say that there is a cause-effect relationship between the two conditions.

### 4.2.4 Epithelial Barrier Hypothesis and Leaky Gut Syndrome (LGS)

The intestinal epithelium is the largest mucosal surface in the body and covers an area of around 300 $m^2$ when the piles of enterocytes are ironed [82]. In the healthy state, tight junction proteins in the intestinal epithelium and the mucus layer form a physical barrier that protects the organism from invading bacteria [83]. Microorganism-derived antigens can enter the systemic circulation with the formation of microdamages in the intestinal enterocyte wall and increased permeability [84]. It is called "leaky gut" [85]. It can trigger an immune response with the introduction of pathogenic antigens into the circulation [86]. With the deterioration of intestinal permeability and entrance of bacteria-derived lipopolysaccharides into the systemic bloodstream, the production of pro-inflammatory cytokines increases with the stimulation of TLRs [62]. As a result of the impairment in intestinal permeability, TLRs are stimulated, and the production of pro-inflammatory cytokines increases due to the bacterial lipopolysaccharides entering the systemic blood circulation.

A low-fiber diet and high glucose intake increase the rates of mucin-degrading bacteria [87, 88]. Antibiotics are another important factor that contributes to the change of microbial composition [89]. Proton pump inhibitors reduce gastric acid barrier and facilitate translocation of oral microbiota pathogens to the gut [90]. Some genetic mutations (NOD2 and XBP1) and environmental stress can cause dysbiosis and Paneth cell dysfunction which has antibacterial activity [91].

In addition, patients with low serum IgA concentrations are highly susceptible to intestinal dysbiosis and allergic and autoimmune diseases (e.g., type 1 diabetes, rheumatoid arthritis, and systemic lupus erythematosus) [92–94].

One of the issues emphasized in recent years is the AP1M2 (encoding the m1B subunit of the AP-1B complex) gene dysfunctions. AP1M2 deficiency was shown to alter the functions of some cytokine receptors (e.g., IL-6, IL-17, tumor necrosis factor) [95]. These abnormalities cause decreased cytokine signaling, disruption of

IgA secretion into the intestinal lumen, and decreased expression of antimicrobial peptides in the intestinal epithelium.

In conclusion, dysbiosis and LGS are seen in mice with AP1M2 deficiency [95]. The importance of AP-1B-mediated functions in systemic immune homeostasis and maintenance of intestinal epithelial integrity is currently being studied.

### 4.2.5 Dysbiosis

The human genome lacks genes encoding enzymes to digest plant-derived polysaccharides. The digestion of these nutrients is possible through the enzymes synthesized by the microbiota [96]. As a result of the digestion of plant-derived polysaccharide fibers, short-chain fatty acids (SCFAs) (these are acetate, butyrate, propionate, lactate) are produced [97]. These SCFAs are absorbed from the colon, enter in the systemic circulation, go to the liver and muscles, and take part in many metabolic functions [97]. A small amount of SCFAs crosses the blood-brain barrier to reach the central nervous system and change neuromodulation [98, 99].

Metabolites of microbiota bacteria are not limited to SCFAs. Metabolites originating from the microbiota and entering the blood circulation have a very important role in neuroimmune disorders and neuroinflammation [100]. For example, the role of metabolites such as serotonin and antioxidant indoxyl sulfate and indole-3-propionic acid synthesized from tryptophan was demonstrated in germ-free animal experiments [101].

It is thought that the normal healthy lumen and its associated immune system physiology change with the change in intestinal bacterial composition, and this change may lead to autoimmune diseases [17, 26].

### 4.2.6 The Role of B Cells

B cells play a crucial role in the dialogue between the mucosal microbiota and the host's immune system due to their capacity to produce IgA antibodies and differentiate into other lymphocytes. The B cell's IgA response is essential for maintaining a healthy microbiota system. Therefore, any change in the IgA response will affect the microbial diversity in the intestinal mucosa and lead to dysbiosis. This dysbiosis can lead to autoimmune inflammation under certain conditions [102].

IgA dimers and J-chain antibody complexes produced in the lamina propria are transported through epithelial cells after binding to the poly-Ig-receptor (pIgR) on the basolateral side of epithelial cells [103]. The IgA response is determined by intestinal bacteria. Pathogenic bacteria induce T-cell-dependent IgA responses, while the vast majority of commensal bacteria induce T-cell-independent IgA responses [104].

In addition to producing antibodies, B cells can also serve as antigen-presenting cells (APC) and can produce significant amounts of cytokines. IL-10-producing B cells, also known as regulatory B cells (Bregs), have the capacity to suppress

autoimmune inflammation. Therefore, gut-resident bacteria and bacterial metabolites can induce abundant IL-10 production from B cells that control anti-inflammatory activity in autoimmunity [105]. In addition, bacterial metabolites can suppress the production of inflammatory cytokines by B cells [106].

While many questions have been cleared about the complex interaction between the microbiota and immune response mediated by B cells in autoimmunity, some fundamental questions still remain. Is intestinal dysbiosis the result of inflammation or one of the environmental triggers of autoimmune inflammation in the susceptible host? Can genetics influence gut microbiota composition by regulating IgA response or regulating epithelial/systemic immune cell function? More research is needed to answer these questions.

### 4.2.7 The Role of Toll-like Receptor (TLR) Ligands

At the molecular level, many arguments suggest that both pathogens and commensal microorganisms primarily interact molecularly with TLRs. Many different infectious agents have TLR ligands. The proof of this is that allergic and autoimmune reactions can be suppressed as a result of systemic administration of TLR ligands [107]. Moreover, it is not necessary for the abovementioned infectious microorganisms to be alive to prevent the onset of autoimmune diseases. The same results can be obtained with bacterial [108] or parasitic materials [77, 78].

Different TLR ligands may have different mechanisms of action depending on the specific receptor. For example, TLR4 ligands act through FoxP3$^+$ regulatory T cells, while TLR3 ligands act through natural killer T (NKT) cells [107].

The same TLR2 desensitization may also play a role in the prevention of experimental allergic encephalomyelitis (EAE) [109]. Administration of low doses of two different TLR2 ligands (Pam2CSK4 and Lipid 654) to formerly encephalitogenic (EAE-inducing) T cells attenuated EAE and reduced the level of TLR2 signaling as well [109, 110]. Interestingly, Lipid 654 is a TLR2 ligand derived from a microbiota commensal and is present in healthy human serum but significantly less in the serum of patients with multiple sclerosis [109, 110]. In another mouse model of EAE, repeated administration of a synthetic TLR7 ligand was reported to significantly reduce disease severity as well as expression of chemokines in the target organ [111].

Studies of children growing up on dairy farms in an LPS-rich environment found a low incidence of allergies, which may be a result of TLR desensitization [112, 113].

### 4.2.8 Autoimmunity

The origin of autoimmunity is long thought to be cross-reacting due to the molecular similarity between antigens on microorganisms and host antigens. Of course, some mimotopes were found in commensal bacteria that were suggested to cause

induction of autoimmune pathogenesis [114, 115] as well as the production of antibodies against host antigens [116].

In addition to the local humoral response in the intestinal mucosa, bacteria and bacterial components entering the systemic bloodstream trigger a systemic antibody reaction beyond the intestinal mucosa. This results in circulating antibodies against bacteria found in the gut [117].

The immune system performs its complex functions through mediators—the most important of which are cytokines—synthesized and secreted by leukocytes and lymphocytes. These mediators in protein structure enable immune cells to communicate with each other [118]. Various cytokines have different effects on immune cells, providing pro-inflammatory or anti-inflammatory stimuli [119]. Pro-inflammatory activity is the primary function of cytokines and they initiate inflammation in body tissues. Cytokines with main pro-inflammatory activity are interleukin (IL)-6, IL-1β, IL-15, IL-17, IL-18, tumor necrosis factor (TNF)-α, and interferon-γ (IFN-γ) [119]. The main anti-inflammatory cytokines that function to inhibit the immune reaction are IL-4, IL10, and IL13 [119, 120]. The imbalance between pro-inflammatory cytokines and anti-inflammatory cytokines may predispose to various immune system-related disorders that adversely affect all body functions.

The main function of the immune system is to find and neutralize pathogenic microorganisms. Molecular extracts of microorganisms (nucleic acids, cell wall components in lipopolysaccharide structure, flagella, etc.) activate immune system cells [17]. Microbiota bacteria come in contact with the immune system through pattern recognition receptors (PRRs). The most important members of PRRs are toll-like receptors (TLRs), as explained in detail above [26].

When PRRs are activated by commensal bacteria, anti-inflammatory cytokines such as interleukin-10 (IL-10) are produced [27]. For example; in humans, *Bifidobacterium infantis* and *Lactobacillus* GG increase IL-10 levels, decrease pro-inflammatory cytokine levels, and restore blood-brain barrier permeability impaired due to inflammation [28]. In addition, beneficial bacteria block the pro-inflammatory process caused by pathogens by activating TLR-2 and TLR-4, [29]. They can also induce prostaglandin synthesis, which provokes the pro-inflammatory process in another way [121]. Psychobiotics play an important role in reducing low-level inflammation by reducing the level of pro-inflammatory cytokines in the systemic circulation.

In recent years, studies in the field of psychoneuroimmunology provided supporting evidence that depression, a prototype neuropsychiatric disease, may also have an autoimmune aspect. Opinions emphasizing the role of autoimmune processes in the etiopathogenesis of depression are increasing [122]. This and similar observations suggest that immune processes and inflammation may play a role in the pathophysiology of depression.

For example, in healthy individuals, the pro-inflammatory cytokine interleukin-6 (IL-6) levels increase after typhoid vaccine, and that may cause depression [123]. Some meta-analysis studies reveal that the levels of pro-inflammatory

cytokines (IL-6, IL1β, tumor necrosis factor alpha, and C-reactive protein) increase in patients with depression and decrease with antidepressant treatments [124, 125].

Is inflammation the cause or the result of depression? This debate is going on for years. However, some recent cohort studies suggest that depression may develop following elevation of pro-inflammatory cytokines with immune stimulation [126, 127]. However, in order to clarify the role of inflammation in depression, it is necessary to differentiate the immune effect on specific symptoms. For example, it was determined that the CRP levels of patients with depression whose somatic symptoms (anergy, sleep, and appetite problems) are at the forefront, rather than psychological symptoms (unhappiness, pessimism, etc.), were increased [128]. A similar situation exists in cancer patients who developed secondary depression due to interferon therapy [129]. It is quite obvious that there is a correlation between depression and immune hyperactivity. However, the evidence obtained in the light of evaluations to establish a causal relationship suggests that an inflammation is likely to be stimulated for an unknown reason before the development of depression. Therefore, the validity of the immune hypothesis in depression is increasing day by day and immune system hyperactivity comes to the forefront as a risk factor for depression.

One of the important areas in the immunological etiopathogenesis of depression is the immune-kynurenine pathway [130]. A common precursor of serotonin and kynurenine is tryptophan [131]. More than 90% of tryptophan is converted to kynurenine through the enzymes indoleamine-2,3-dioxygenase (IDO) found in all somatic cells and tryptophan-2,3-dioxygenase (TDO) found only in hepatocytes [132, 133]. IDO is in two different configurations, IDO1 and IDO2 [134]. When systemic inflammation occurs, IDO1 and TDO activity rate increases. Pro-inflammatory cytokines and glucocorticoids, molecular precursors of systemic inflammation, convert tryptophan to kynurenine by strongly stimulating both the IDO1 and TDO pathways [133]. Metabolites of kynurenine (kynurenic acid and quinolinic acid) have a stimulating effect on N-methyl-D-aspartate (NMDA) and alpha-7 nicotinic cholinergic receptors [135]. Kynurenic acid exhibits anti-inflammatory and neuroprotective properties, while quinolinic acid exhibits excitotoxic properties [136, 137].

In order to put the immune-kynurenine pathway in its place in the etiology chain of depression, we also need to address the gut-brain axis. When the intestinal microbiota composition is disrupted and dysbiosis develops, serotonin synthesis from tryptophan in the intestines decreases [130]. In addition, lipopolysaccharides that enter the systemic circulation due to leaky gut can stimulate the immune system and trigger low-grade inflammation [17]. Under these conditions, the production of serotonin from tryptophan is further reduced by the effect of additional inflammation added to the cycle, and tryptophan catabolism shifts from serotonin to the kynurenine pathway. This cyclical system plays a role in the etiopathogenesis of depression and may feed the autoimmune reaction [17].

## 4.3 Novel Therapeutic Intervention

### 4.3.1 Engineering the Gut Microbiota

Is it possible to carry out engineering studies to restore the impaired microbiota balances? Can obtaining more efficient bacterial metabolites and creating stronger commensal bacterial strains through engineered bacteria provide further therapeutic benefits?

For any disease, when long-term administration of a compound with therapeutic properties is required, engineered bacteria with a high potential can be used for colonization [138]. In addition, this method can reduce the off-target effects of a compound by the slow release of the targeted beneficial metabolite locally (in the small intestine or colon).

To date, products designed to be produced by therapeutic bacteria included small molecules, vaccine antigens, enzymes, interleukins, and antibodies. Any type of bacteria found in the human gut can theoretically be engineered. Because the composition of the gut microbiota varies both longitudinally (e.g., ileum vs. colon) and transversely (e.g., intestinal mucus vs. lumen), bacteria localized to the most suitable target site for modification shall be selected [139].

However, the technical process appears to be directed by practical considerations such as ease of genetic manipulation, large-scale culture, and low toxicity. Therefore, the most commonly used genetically modified strains are lactic acid bacteria and *Escherichia coli* strains [140]. These strains are being tested in the treatment of inflammatory bowel diseases, cancer, diabetes, cardiometabolic syndrome, and phenylketonuria [141].

### 4.3.2 Personalized Nutrition

The impact of diet on health and disease has been studied for decades and includes a variety of pediatric and adult disorders, particularly diseases such as obesity, type 2 diabetes (T2D), myocardial infarction, stroke, and nonalcoholic hepatosteatosis [142]. Indeed, changes in our dietary habits over the past few decades have been associated with a large increase in the prevalence of these diseases.

At the same time, it is clearly observed that the prevalence of certain diseases such as T2D and dyslipidemia—which were known as adult age group problems in the past that leads to cardiovascular disorders later on if not addressed early—and obesity are increasing in children [142].

Today, obesity has become a global epidemic, and decades of nutritional advice from health organizations have not alleviated this epidemic. The reason for this is that general nutritional recommendations do not apply to individuals. Several recent studies show that one diet paradigm may not suit all [142]. Responses to diet are heterogeneous among different individuals, and this heterogeneity is driven by our individual genetic makeup and gut microbiota composition [142, 143]. This view

raised a growing interest in the role and potential of personalized nutritional interventions.

Technological advances allow not only a more complex understanding of the mechanisms that cause disease, but also a more comprehensive understanding of the underlying mechanisms that act to cure disease [142, 143]. Increasing knowledge about the personalized effects of dietary components suggests that diet may have a potential role in the prevention and treatment of autoimmune and immune system-mediated neuropsychiatric diseases, especially metabolic diseases.

### 4.3.3 Probiotics and Prebiotics

Bacteria colonizing the microbiota also need food like other living things. Foods or substances that allow certain intestinal bacteria to grow more in number are called prebiotics [12]. For example, *Bacteroides fragilis* and *Faecalibacterium prausnitzii* synthesize SCFAs (acetate, butyrate, propionate) from fiber. For these bacteria, fiber has prebiotic properties and these metabolites have an anti-inflammatory effect [144, 145].

Oral or rectal administration of a commensal microorganism to regulate the microbiota composition is called probiotic therapy [146]. There are many publications stating that probiotics are beneficial in the treatment of depression as a disease with autoimmune etiopathogenesis [147, 148]. This beneficial effect is probably due to the regulation of the immune system and anti-inflammatory activity.

In a study compiling ten randomized controlled trials (RCT) published between 1990 and 2016, it was found that probiotics have a positive effect on depression and anxiety symptoms [149]. However, in a study conducted on 18,019 people between 2005 and 2012, no relationship was found between probiotics and low depression rates [150]. In another RCT, it was determined that 8-week-long *Lactobacillus helveticus* and *Bifidobacterium longum* application was not effective on depressive symptoms [151].

There is a widespread opinion that prebiotic and probiotic supplements have positive effects on the immune system and may play a role in the control of autoimmune diseases by exhibiting anti-inflammatory activity in this direction. However, more clear evidence is needed between prebiotics/probiotics and immune mechanisms in order to be included in the standard treatment modalities.

### 4.3.4 Fecal Microbiota Transplantation (FMT)

FMT is the process of suspending stool from a healthy donor and transporting it to the intestines of the sick person [26]. Although the history of FMT dates back centuries ago in traditional medicine, it was first applied in 1958 in modern medicine. It was reported that cases of pseudomembranous enterocolitis caused by *Clostridium difficile* were successfully treated with this method [152]. A second case series of Clostridium difficile infection (CDI) treated with FMT was reported

23 years after the first one [153]. After these publications, the mechanism of action and safety of FMT were researched extensively.

The main indications for use of FMT are CDI and inflammatory bowel diseases such as ulcerative colitis and Crohn's disease. It is effective on irritable bowel diseases and psychiatric symptoms [154]. The main purpose of FMT application is to repair and regulate the intestinal microbiota that has become highly dysbiotic.

Although the general medical condition of the patients administered stool transplantation is quite poor, FMT is a safe practice. In previous publications, very low rates and probabilities of serious side effects were reported [155–158]. Because of the huge amount of knowledge accumulated over the last 40 years, FMT appears to be an increasingly prominent modality in the treatment of autoimmune disorders [159].

### 4.3.5 Vaccination

Is the vaccine option reasonable and reasonable in the prevention or treatment of autoimmune diseases? Rook and colleagues speculated in 2012 whether a vaccine could be developed to treat depression [160]. This idea, like every innovative idea, seems to be found strange by the scientific world, because there is no vaccine studied for the treatment of depression in the literature [161]. In fact, considering the low rate of treatment with standard antidepressant drugs and the high rate of relapse, it is very important to have a vaccine option especially for the prevention of recurrent depression, because, according to the rapidly increasing literature, the immune component in depression is quite obvious.

Vaccines are generally developed for a certain infectious disease and are treatments applied to immunize against the pathogenic microorganism that is the cause of that disease [162]. In other words, in order to develop a vaccine, it is necessary to determine the pathogen that plays a role in the etiopathogenesis of the disease. The same route shall also be used to develop a vaccine against depression.

In this direction, those in the intestinal microbiota shall come first among the targeted microorganisms. A vaccine containing bacteria and their metabolites that enter the systemic circulation due to leaky gout may have a chance of success. In this direction, for example, it was determined that IgA and IgM levels for LPS of gram-negative enterococci (Proteobacteria) were increased in depression cases [163]. In addition, fecal microbiota analyses of depressed patients showed high Bacteroidetes levels [164]. However, there are studies showing that it is possible to transmit depression [165, 166]. If depression and many other autoimmune diseases are caused by certain microorganism, and after these are clearly identified, we can be hopeful for the development of a vaccine in the future.

## 4.4 Conclusion

It has been 350 years since the Dutch scientist Antonie Philips van Leeuwenhoek, who is considered the "father of microbiology," discovered bacteria. All this time, science has tried to understand the relationship between unicellular and multicellular organisms. Today, we know that this interaction has existed for millions of years and that eukaryotes coevolved with prokaryotes. Commensal bacteria living in the microbiome—so to speak—train our immune system. Any disruptions in this education may pave the way for autoimmune diseases. The area where the microbiota-host interaction is most intensely studied is the gut-brain axis. There is comprehensive and up-to-date information about the role of the microbiota in the formation of inflammation and the neuropsychiatric disorders caused by neuroinflammation in the other chapters of this book. In this chapter, the role of the microbiome in the formation of autoimmunity and new treatment options emerged on this basis are examined. Although these new therapeutic approaches (prebiotics, probiotics, fecal microbiota transplantation, bacterial engineering, and vaccination) are still in their infancy, it seems possible that they can be applied more specifically and safely in the future.

## References

1. O'Brien S, Goedert J. HIV causes AIDS: Koch's postulates fulfilled. Curr Opin Immunol. 1996;8:613–8.
2. Hempelmann E, Krafts K. Bad air, amulets and mosquitoes: 2,000 years of changing perspectives on malaria. Malar J. 2013;12:232.
3. Mackowiak PA. Recycling Metchnikoff: probiotics, the intestinal microbiome and the quest for long life. Front Public Health. 2013;1:52.
4. Phillips J. The treatment of melancholia by the lactic acid bacillus. J Mental Sci. 1910;56:422–30.
5. Strachan DP. Hay fever, hygiene, and household size. BMJ. 1989;299:1259–60.
6. Rook GA. Review series on helminths, immune modulation and the hygiene hypothesis: the broader implications of the hygiene hypothesis. Immunology. 2009;126:3–11.
7. Sender R, Fuchs S, Milo R. Revised estimates for the number of human and bacteria cells in the body. PLoS Biol. 2016;14:e1002533.
8. Zhu B, Wang X, Li L. Human gut microbiome: the second genome of human body. Protein Cell. 2010;1:718–25.
9. Stilling RM, Dinan TG, Cryan JF. Microbial genes, brain & behaviour—epigenetic regulation of the gut-brain axis. Genes Brain Behav. 2014;13:69–86.
10. Lozupone CA, Stombaugh JI, Gordon JI, Jansson JK, Knight R. Diversity, stability and resilience of the human gut microbiota. Nature. 2012;489:220–30.
11. Whitman WB, Coleman DC, Wiebe WJ. Prokaryotes: the unseen majority. Proc Natl Acad Sci U S A. 1998;95:6578–83.
12. Evrensel A, Ünsalver BÖ, Ceylan ME. Psychobiotics. Adv Exp Med Biol. 2019;1192:565–81.
13. Rieder R, Wisniewski PJ, Alderman BL, Campbell SC. Microbes and mental health: a review. Brain Behav Immun. 2017;66:9–17.
14. Abisado RG, Benomar S, Klaus JR, Dandekar AA, Chandler JR. Bacterial quorum sensing and microbial community interactions. MBio. 2018;9:e02331–17.

15. Dinan TG, Stilling RM, Stanton C, Cryan JF. Collective unconscious: how gut microbes shape human behavior. J Psychiatr Res. 2015;63:1–9.
16. Branton WG, Ellestad KK, Maingat F. Brain microbial populations in HIV/AIDS: α-proteobacteria predominate independent of host immune status. PLoS One. 2013;8:e54673.
17. Evrensel A, Ceylan ME. The gut-brain Axis: the missing link in depression. Clin Psychopharmacol Neurosci. 2015;13:239–44.
18. Davey KJ, O'Mahony SM, Schellekens H, et al. Olanzapine induced weight gain in the rat: impact on inflammatory, metabolic and microbiota parameters. Psychopharmacology. 2013;221:155–69.
19. Bailey MT, Coe CL. Maternal separation disrupts the integrity of the intestinal microflora in infant rhesus monkeys. Dev Psychobiol. 1999;35:146–55.
20. O'Mahony SM, Marchesi JR, Scully P, et al. Early life stress alters behavior, immunity, and microbiota in rats: implications for irritable bowel syndrome and psychiatric illnesses. Biol Psychiatry. 2009;65:263–7.
21. Bailey MT, Dowd SE, Galley JD, Hufnagle AR, Allen RG, Lyte M. Exposure to a social stressor alters the structure of the intestinal microbiota: implications for stressor-induced immunomodulation. Brain Behav Immun. 2011;25:397–407.
22. Desbonnet L, Clarke G, Traplin A, et al. Gut microbiota depletion from early adolescence in mice: implications for brain and behavior. Brain Behav Immun. 2015;48:165–73.
23. Macedo D, Filho AJMC, Soares de Sousa CN. Antidepressants, antimicrobials or both? Gut microbiota dysbiosis in depression and possible implications of the antimicrobial effects of antidepressant drugs for antidepressant effectiveness. J Affect Disord. 2017;208:22–32.
24. Dash S, Clarke G, Berk M, Jacka FN. The gut microbiome and diet in psychiatry: focus on depression. Curr Opin Psychiatry. 2015;28:16.
25. David LA, Maurice CF, Carmody RN, et al. Diet rapidly and reproducibly alters the human gut microbiome. Nature. 2014;505:559–63.
26. Evrensel A, Ceylan ME. Fecal microbiota transplantation and its usage in neuropsychiatric disorders. Clin Psychopharmacol Neurosci. 2016;14:231–7.
27. Chu H, Mazmanian SK. Innate immune recognition of the microbiota promotes host-microbial symbiosis. Nat Immunol. 2013;14:668–75.
28. O'Mahony L, McCarthy J, Kelly P, et al. Lactobacillus and bifidobacterium in irritable bowel syndrome: symptom responses and relationship to cytokine profiles. Gastroenterology. 2005;128:541–51.
29. Zhou W, Lv H, Li MX. Protective effects of bifidobacteria on intestines in newborn rats with necrotizing enterocolitis and its regulation on TLR2 and TLR4. Genet Mol Res. 2015;14:11505–14.
30. Ganal-Vonarburg SC, Hornef MW, Macpherson AJ. Microbial–host molecular exchange and its functional consequences in early mammalian life. Science. 2020;368:604–7.
31. Aagaard K, Ma J, Antony KM, Ganu R, Petrosino J, Versalovic J. The placenta harbors a unique microbiome. Sci Transl Med. 2014;6:237ra65.
32. Rautava S, Collado MC, Salminen S, Isolauri E. Probiotics modulate host- microbe interaction in the placenta and fetal gut: a randomized, double-blind, placebo-controlled trial. Neonatology. 2012;102:178–84.
33. Stout MJ, Conlon B, Landeau M, et al. Identification of intracellular bacteria in the basal plate of the human placenta in term and preterm gestations. Am J Obstet Gynecol. 2013;208(3):226.e1–7.
34. Jiménez E, Fernández L, Marıń ML, et al. Isolation of commensal bacteria from umbilical cord blood of healthy neonates born by cesarean section. Curr Microbiol. 2005;51:270–4.
35. Jiménez E, Marıń ML, Martıń R, et al. Is meconium from healthy newborns actually sterile? Res Microbiol. 2008;159:187–93.
36. Collado MC, Rautava S, Aakko J. Human gut colonization may be initiated in utero by distinct microbial communities in the placenta and amniotic fluid. Sci Rep. 2016;6:23129.

37. Bassols J, Serino M, Carreras-Badosa G, et al. Gestational diabetes is associated with changes in placental microbiota and microbiome. Pediatr Res. 2016;80:777–84.
38. Antony KM, Ma J, Mitchell KB, Racusin DA, Versalovic J, Aagaard K. The preterm placental microbiome varies in association with excess maternal gestational weight gain. Am J Obstet Gynecol. 2015;212:653.e1–e16.
39. Zheng J, Xiao X, Zhang Q, Mao L, Yu M, Xu J. The placental microbiome varies in association with low birth weight in full-term neonates. Nutrients. 2015;7:6924–37.
40. Kliman HJ. Comment on "the placenta harbors a unique microbiome". Sci Trans Med. 2014;6: 254le4.
41. de Goffau MC, Lager S, Sovio U, Gaccioli F, Cook E, Peacock SJ, et al. Human placenta has no microbiome but can contain potential pathogens. Nature. 2019;572:329–34.
42. Salter SJ, Cox MJ, Turek EM, et al. Reagent and laboratory contamination can critically impact sequence- based microbiome analyses. BMC Biol. 2014;12:87.
43. Olomu IN, Pena-Cortes LC, Long RA, et al. Elimination of "kitome" and "splashome" contamination results in lack of detection of a unique placental microbiome. BMC Microbiol. 2020;20:157.
44. Gevers D, Kugathasan S, Denson LA, et al. The treatment-naive microbiome in new-onset crohn's disease. Cell Host Microbe. 2014;15:382–92.
45. Frank DN, St. Amand AL, Feldman RA, Boedeker EC, Harpaz N, Pace NR. Molecular-phylogenetic characterization of microbial community imbalances in human inflammatory bowel diseases. Proc Natl Acad Sci. 2007;104:13780–5.
46. Ni J, Shen T-CD, Chen EZ, et al. A role for bacterial urease in gut dysbiosis and Crohn's disease. Sci Trans Med. 2017;15:eaah6888.
47. Kostic Aleksandar D, Chun E, Robertson L, et al. Fusobacterium nucleatum potentiates intestinal tumorigenesis and modulates the tumor-immune microenvironment. Cell Host Microbe. 2013;14:207–15.
48. Nejman D, Livyatan I, Fuks G, et al. The human tumor microbiome is composed of tumor type–specific intracellular bacteria. Science. 2020;368:973–80.
49. Kuperman A, Zimmerman A, Hamadia S, et al. Deep microbial analysis of multiple placentas shows no evidence for a placental microbiome. BJOG Int J Obstet Gynaecol. 2020;127:159–69.
50. Lauder AP, Roche AM, Sherrill-Mix S, et al. Comparison of placenta samples with contamination controls does not provide evidence for a distinct placenta microbiota. Microbiome. 2016;4:29.
51. Theis KR, Romero R, Winters AD, et al. Does the human placenta delivered at term have a microbiota? Results of cultivation, quantitative real-time PCR, 16s rRNA gene sequencing, and metagenomics. Am J Obstet Gynecol. 2019;220(3):e1-267–e39.
52. Li Y, Toothaker JM, Ben-Simon S, et al. In utero human intestine Harbors unique metabolome, including bacterial metabolites. JCI Insight. 2020;5:e138751.
53. Rackaityte E, Halkias J, Fukui EM, et al. Viable bacterial colonization is highly limited in the human intestine in utero. Nat Med. 2020;26:599–607.
54. Rackaityte E, Halkias J, Fukui EM, et al. Corroborating evidence refutes batch effect as explanation for fetal bacteria. Microbiome. 2021;9:10.
55. de Goffau MC, Charnock-Jones DS, Smith GCS, Parkhill J. Batch effects account for the main findings of an in utero human intestinal bacterial colonization study. Microbiome. 2021;9:6.
56. Olszak T, An D, Zeissig S, et al. Microbial exposure during early life has persistent effects on natural killer T cell function. Science. 2012;336:489–93.
57. McKernan DP, Dennison U, Gaszner G, Cryan JF, Dinan TG. Enhanced peripheral toll-like receptor responses in psychosis: further evidence of a pro-inflammatory phenotype. Transl Psychiatry. 2011;1:e36.
58. McCusker RH, Kelley KW. Immune-neural connections: how the immune system's response to infectious agents influences behavior. J Exp Biol. 2013;216:84–98.

59. Dinan TG, Quigley EM. Probiotics in the treatment of depression: science or science fiction? Aust N Z J Psychiatry. 2011;45:1023–5.
60. Carvalho FA, Aitken JD, Vijay-Kumar M, Gewirtz AT. Toll-like receptor-gut microbiota interactions: perturb at your own risk! Annu Rev Physiol. 2012;74:177–98.
61. Lucas K, Maes M. Role of the toll like receptor (TLR) radical cyclein chronic inflammation: possible treatments targeting the TLR4 pathway. Mol Neurobiol. 2013;48:190–204.
62. Ait-Belgnaoui A, Durand H, Cartier C, et al. Prevention of gut leakiness by intestinal microbiota modulation leads to attenuated HPA response to an acute psychological stress in rats. Psychoneuroendocrinology. 2012;37:1885–95.
63. Smythies LE, Smythies JR. Microbiota, the immune system, black moods and the brain melancholia updated. Front Hum Neurosci. 2014;8:720.
64. Costello EK, Stagaman K, Dethlefsen L, Bohannan J, Relman DA. The application of ecological theory toward an understanding of the human microbiome. Science. 2012;336: 1255–62.
65. Strachan DP. Family size, infection and atopy: the first decade of the "hygiene hypothesis". Thorax. 2000;55:S2–10.
66. Bach JF. The effect of infections on susceptibility to autoimmune and allergic diseases. N Engl J Med. 2002;347:911–20.
67. Greenwood BM, Herrick EM, Voller A. Suppression of autoimmune disease in NZB and (NZB x NZW) F1 hybrid mice by infection with malaria. Nature. 1970;226:266–7.
68. Patterson CC, Gyurus E, Rosenbauer J, et al. Trends in childhood type 1 diabetes incidence in Europe during 1989-2008: evidence of non-uniformity over time in rates of increase. Diabetologia. 2012;55:2142–7.
69. Negrato CA, Lauris JRP, Saggioro IB, et al. Increasing incidence of type 1 diabetes between 1986 and 2015 in Bauru, Brazil. Diabetes Res Clin Pract. 2017;127:198–204.
70. Karvonen M, Pitkaniemi J, Tuomilehto J. The onset age of type 1 diabetes in Finnish children has become younger. The Finnish childhood diabetes registry group. Diabetes Care. 1999;22: 1066–70.
71. Almeida MC, Lima GS, Cardoso LS. The effect of antihelminthic treatment on subjects with asthma from an endemic area of schistosomiasis: a randomized, double-blinded, and placebo-controlled trial. J Parasitol Res. 2012;2012:296856.
72. Pelucchi C, Chatenoud L, Turati F, et al. Probiotics supplementation during pregnancy or infancy for the prevention of atopic dermatitis: a meta-analysis. Epidemiology. 2012;23:402–14.
73. Charabati M, Donkers SJ, Kirkland MC, Osborne LC. A critical analysis of helminth immunotherapy in multiple sclerosis. Mult Scler. 2020;26:1448–58.
74. Dixit A, Tanaka A, Greer JM, Donnelly S. Novel therapeutics for multiple sclerosis designed by parasitic worms. Int J Mol Sci. 2017;18:2141.
75. Larson JD, Thurman JM, Rubtsov AV, et al. Murine gammaherpesvirus 68 infection protects lupus-prone mice from the development of autoimmunity. Proc Natl Acad Sci U S A. 2012;109:E1092–100.
76. Olia A, Shimokawa C, Imai T, Suzue K, Hisaeda H. Suppression of systemic lupus erythematosus in NZBWF1 mice infected with hymenolepis microstoma. Parasitol Int. 2020;76:102057.
77. Wu Z, Wang L, Tang Y, Sun X. Parasite-derived proteins for the treatment of allergies and autoimmune diseases. Front Microbiol. 2017;8:2164.
78. Harnett MM, Harnett W. Can parasitic worms cure the modern World's ills? Trends Parasitol. 2017;33:694–705.
79. Shimokawa C, Kato T, Takeuchi T, et al. CD8 (+) regulatory T cells are critical in prevention of autoimmune-mediated diabetes. Nat Commun. 2020;11:1922.
80. Tang CL, Zou JN, Zhang RH, Liu ZM, Mao CL. Helminths protect against type 1 diabetes: effects and mechanisms. Parasitol Res. 2019;118:1087–94.

81. Donskow-Łysoniewska K, Krawczak K, Machcińska M, Głaczyńska M, Doligalska M. Effects of intestinal nematode treatment on CD11b activation state in an EAE mouse model of multiple sclerosis. Immunobiology. 2019;224:817–26.
82. Helander HF, Fändriks L. Surface area of the digestive tract—revisited. Scand J Gastroenterol. 2014;49:681–9.
83. Borre YE, O'Keeffe GW, Clarke G, Stanton C, Dinan TG, Cryan JF. Microbiota and neurodevelopmental windows: implications for brain disorders. Trends Mol Med. 2014;20: 509–18.
84. Hornig M. The role of microbes and autoimmunity in the pathogenesis of neuropsychiatric illness. Curr Opin Rheumatol. 2013;25:488–95.
85. Evrensel A, Ceylan ME. Microbiome: the missing link in neuropsychiatric disorders. EMJ Innov. 2017;1:83–8.
86. Fetissov SO, Déchelotte P. The new link between gut–brain axis and neuropsychiatric disorders. Curr Opin Clin Nutr Metab Care. 2011;14:477–82.
87. Desai MS, Seekatz AM, Koropatkin NM, Kamada N, Hickey CA, Wolter M, et al. A dietary fiber deprived gut microbiota degrades the colonic mucus barrier and enhances pathogen susceptibility. Cell. 2016;167:1339–53.e21.
88. Khan S, Waliullah S, Godfrey V, et al. Dietary simple sugars alter microbial ecology in the gut and promote colitis in mice. Sci Transl Med. 2020;12:eaay6218.
89. Feng Y, Huang Y, Wang Y, Wang P, Song H, Wang F. Antibiotics induced intestinal tight junction barrier dysfunction is associated with microbiota dysbiosis, activated NLRP3 inflammasome and autophagy. PLoS One. 2019;14:e0218384.
90. Imhann F, Bonder MJ, Vila AV, et al. Proton pump ınhibitors affect the gut microbiome. Gut. 2016;65:740–8.
91. Salzman NH, Bevins CL. Dysbiosis—a consequence of paneth cell dysfunction. Semin Immunol. 2013;25:334–41.
92. Berbers RM, Franken IA, Leavis HL. Immunoglobulin a and microbiota in primary immunodeficiency diseases. Curr Opin Allergy Clin Immunol. 2019;19:563–70.
93. Jorgensen GH, Gardulf A, Sigurdsson MI, et al. Clinical symptoms in adults with selective IgA deficiency: a case-control study. J Clin Immunol. 2013;33:742–7.
94. Ludvigsson JF, Neovius M, Hammarström L. Association between IgA deficiency & other autoimmune conditions: a population-based matched cohort study. J Clin Immunol. 2014;34: 444–51.
95. Takahashi D, Hase K, Kimura S, Nakatsu F, Ohmae M. The epithelia-specific membrane trafficking factor AP-1B controls gut immune homeostasis in mice. Gastroenterology. 2011;141:621–32.
96. Qin J, Li R, Raes J, et al. A human gut microbial gene catalogue established by metagenomic sequencing. Nature. 2010;464:59–65.
97. Tan J, McKenzie C, Potamitis M, Thorburn AN, Mackay CR, Macia L. The role of short-chain fatty acids in health and disease. Adv Immunol. 2014;121:91–119.
98. Lei E, Vacy K, Boon WC. Fatty acids and their therapeutic potential in neurological disorders. Neurochem Int. 2016;95:75–84.
99. Erny D, Hrabě de Angelis AL, Jaitin D, et al. Host microbiota constantly control maturation and function of microglia in the CNS. Nat Neurosci. 2015;18:965–77.
100. Morris G, Berk M, Carvalho A, et al. The role of the microbial metabolites including tryptophan catabolites and short chain fatty acids in the pathophysiology of immune-inflammatory and neuroimmune disease. Mol Neurobiol. 2017;54:4432–51.
101. Yano JM, Yu K, Donaldson GP, et al. Indigenous bacteria from the gut micro-biota regulate host serotonin biosynthesis. Cell. 2015;161:264–76.
102. Okai S, Usui F, Ohta M, et al. Intestinal IgA as a modulator of the gut microbiota. Gut Microbes. 2017;8:486–92.
103. Pabst O, Slack E. IgA and the intestinal microbiota: the importance of being specific. Mucosal Immunol. 2020;13:12–21.

104. Bunker JJ, Flynn TM, Koval C, et al. Innate and adaptive humoral responses coat distinct commensal bacteria with immunoglobulin A. Immunity. 2015;43:541–53.
105. Mu Q, Edwards MR, Swartwout BK, et al. Gut microbiota and bacterial dna suppress autoimmunity by stimulating regulatory B cells in a murine model of lupus. Front Immunol. 2020;11:593353.
106. Huang J, Pearson JA, Peng J, et al. Gut microbial metabolites alter IgA immunity in type 1 diabetes. JCI Insight. 2020;5:e135718.
107. Aumeunier A, Grela F, Ramadan, et al. Systemic toll-like receptor stimulation suppresses experimental allergic asthma and autoimmune diabetes in NOD mice. PLoS One. 2010;5: e11484.
108. Alyanakian MA, Grela F, Aumeunier A, et al. Transforming growth factor-beta and natural killer T-cells are involved in the protective effect of a bacterial extract on type 1 diabetes. Diabetes. 2006;55:179–85.
109. Wasko NJ, Nichols F, Clark RB. Multiple sclerosis, the microbiome, TLR2, and the hygiene hypothesis. Autoimmun Rev. 2020;19:102430.
110. Anstadt EJ, Fujiwara M, Wasko N, Nichols F, Clark RB. TLR tolerance as a treatment for central nervous system autoimmunity. J Immunol. 2016;197:2110–8.
111. Hayashi T, Yao S, Crain B, Chan M, Tawatao RI, Gray C, et al. Treatment of autoimmune inflammation by a TLR7 ligand regulating the innate immune system. PLoS One. 2012;7: e45860.
112. von Mutius E, Vercelli D. Farm living: effects on childhood asthma and allergy. Nat Rev Immunol. 2010;10:861–8.
113. Schuijs MJ, Willart MA, Vergote K, et al. Farm dust and endotoxin protect against allergy through A20 induction in lung epithelial cells. Science. 2015;349:1106–10.
114. Greiling TM, Dehner C, Chen X, et al. Commensal orthologs of the human autoantigen Ro60 as triggers of autoimmunity in lupus. Sci Transl Med. 2018;10:eaan2306.
115. Ruff WE, Dehner C, Kim WJ, et al. Pathogenic autoreactive T and B cells cross-react with mimotopes expressed by a common human gut commensal to trigger autoimmunity. Cell Host Microbe. 2019;26(100–113):E8.
116. Sun W, Gudi RR, Johnson BM, Vasu C. Abundance and nuclear antigen reactivity of intestinal and fecal immunoglobulin a in lupus-prone mice at younger ages correlate with the onset of eventual systemic autoimmunity. Sci Rep. 2020;10:1425.
117. Zeng MY, Cisalpino D, Varadarajan S. Gut microbiota-induced immunoglobulin G controls systemic infection by symbiotic bacteria and pathogens. Immunity. 2016;44:647–58.
118. Reichenberg A, Yirmiya R, Schuld A, et al. Cytokine-associated emotional and cognitive disturbances in humans. Arch Gen Psychiatry. 2001;58:445–52.
119. Alcami A. Viral mimicry of cytokines, chemokines and their receptors. Nat Rev Immunol. 2003;3:36–50.
120. Mechawar N, Savitz J. Neuropathology of mood disorders: do we see the stigmata of inflammation? Transl Psychiatry. 2016;6:e946.
121. Felger JC, Lotrich FE. Inflammatory cytokines in depression: neurobiological mechanisms and therapeutic implications. Neuroscience. 2013;246:199–229.
122. Shin C, Kim YK. Autoimmunity in microbiome-mediated diseases and novel therapeutic approaches. Curr Opin Pharmacol. 2019;49:34–42.
123. Harrison NA, Brydon L, Walker C. Inflammation causes mood changes through alterations in subgenual cingulate activity and mesolimbic connectivity. Biol Psychiatry. 2009;66:407–14.
124. Haapakoski R, Mathieu J, Ebmeier KP, Alenius H, Kivimäki M. Cumulative meta-analysis of interleukins 6 and 1β, tumour necrosis factor α and C-reactive protein in patients with major depressive disorder. Brain Behav Immun. 2015;49:206–15.
125. Goldsmith DR, Rapaport MH, Miller BJ. A meta-analysis of blood cytokine network alterations in psychiatric patients: comparisons between schizophrenia, bipolar disorder and depression. Mol Psychiatry. 2016;21:1696–709.

126. Khandaker GM, Pearson RM, Zammit S, Lewis G, Jones PB. Association of serum interleukin 6 and C-reactive protein in childhood with depression and psychosis in young adult life: a population-based longitudinal study. JAMA Psychiat. 2014;71:1121–8.
127. Zalli A, Jovanova O, Hoogendijk WJ, Tiemeier H, Carvalho LA. Low-grade inflammation predicts persistence of depressive symptoms. Psychopharmacology. 2016;233:1669–78.
128. Jokela M, Virtanen M, Batty GD, Kivimäki M. Inflammation and specific symptoms of depression. JAMA Psychiat. 2016;73:87–8.
129. Capuron L, Gumnick JF, Musselman DL, et al. Neurobehavioral effects of interferon-alpha in cancer patients: phenomenology and paroxetine responsiveness of symptom dimensions. Neuropsychopharmacology. 2002;26:643–52.
130. Evrensel A, Ünsalver BÖ, Ceylan ME. Immune-kynurenine pathways and the gut microbiota-brain axis in anxiety disorders. Adv Exp Med Biol. 2020;1191:155–67.
131. Palego L, Betti L, Rossi A, Giannaccini G. Tryptophan biochemistry: structural, nutritional, metabolic, and medical aspects in humans. J Amino Acids. 2016;2016:8952520.
132. Clarke G, McKernan DP, Gaszner G. A distinct profile of tryptophan metabolism along the kynurenine pathway downstream of toll-like receptor activation in irritable bowel syndrome. Front Pharmacol. 2012;3:90.
133. O'Mahony SM, Clarke G, Borre YE, Dinan TG, Cryan JF. Serotonin, tryptophan metabolism and the brain-gut-microbiome axis. Behav Brain Res. 2015;277:32–48.
134. Fatokun AA, Hunt NH, Ball HJ. Indoleamine 2,3-dioxygenase 2 (IDO2) and the kynurenine pathway: characteristics and potential roles in health and disease. Amino Acids. 2013;45:1319–29.
135. Forrest CM, Youd P, Kennedy A, et al. Purine, kynurenine, neopterin and lipid peroxidation levels in inflammatory bowel disease. J Biomed Sci. 2002;9:436–42.
136. Kaszaki J, Erces D, Varga G. Kynurenines and intestinal neurotransmission: the role of N-methyl-D-aspartate receptors. J Neural Transm. 2012;119:211–23.
137. Stone TW, Darlington LG. The kynurenine pathway as a therapeutic target in cognitive and neurodegenerative disorders. Br J Pharmacol. 2013;169:1211–27.
138. Pinero-Lambea C, Ruano-Gallego D, Fernandez LA. Engineered bacteria as therapeutic agents. Curr Opin Biotechnol. 2015;35:94–102.
139. Sekirov I, Russell SL, Antunes LCM, Finlay BB. Gut microbiota in health and disease. Physiol Rev. 2010;90:859–904.
140. Behnsen J, Deriu E, Sassone-corsi M, Raffatellu M. Probiotics: properties, examples, and specific applications. Cold Spring Harb Perspect Med. 2013;3:a010074.
141. Dosoky NS, May-Zhang LS, Davies SS. Engineering the gut microbiota to treat chronic diseases. Appl Microbiol Biotechnol. 2020;104:7657–71.
142. Ordovas JM, Ferguson LR, Tai ES, et al. Personalised nutrition and health. BMJ. 2018;361:bmjk2173.
143. Valdes AM, Walter J, Segal E, et al. Role of the gut microbiota in nutrition and health. BMJ. 2018;361:k2179.
144. Macfarlane S, Macfarlane GT. Regulation of short-chain fatty acid production. Proc Nutr Soc. 2003;62:67–72.
145. Bollrath J, Powrie F. Feed your tregs more fiber science. Immunology. 2013;341:463–4.
146. Khanna S, Tosh PK. A clinician's primer on the role of the microbiome in human health and disease. Mayo Clin Proc. 2014;89:107–14.
147. Messaoudi M, Lalonde R, Violle N, et al. Assessment of psychotropic-like properties of a probiotic formulation (lactobacillus helveticus R0052 and Bifidobacterium longum R0175) in rats and human subjects. Br J Nutr. 2011;105:755–64.
148. Messaoudi M, Violle N, Bisson JF, Desor D, Javelot H, Rougeot C. Beneficial psychological effects of a probiotic formulation (Lactobacillus helveticus R0052 and Bifidobacterium longum R0175) in healthy human volunteers. Gut Microbes. 2011;2:256–61.

149. Pirbaglou M, Katz J, de Souza RJ, Stearns JC, Motamed M, Ritvo P. Probiotic supplementation can positively affect anxiety and depressive symptoms: a systematic review of randomized controlled trials. Nutr Res. 2016;36:889–98.
150. Cepeda MS, Katz EG, Blacketer C. Microbiome-gut-brain axis: probiotics and their association with depression. J Neuropsychiatry Clin Neurosci. 2017;29:39–44.
151. Romijn AR, Rucklidge JJ, Kuijer RG, Frampton CA. Double-blind, randomized, placebo-controlled trial of lactobacillus helveticus and Bifidobacterium longum for the symptoms of depression. Aust N Z J Psychiatry. 2018;51:810–21.
152. Eiseman B, Silen W, Bascom GS, Kauvar AJ. Fecal enema as an adjunct in the treatment of pseudomembranous enterocolitis. Surgery. 1958;44:854–9.
153. Bowden TA, Mansberger AR, Lykins LE. Pseudomembranous enterocolitis: mechanism of restoring floral homeostasis. Am Surg. 1981;47:178–83.
154. Kilinçarslan S, Evrensel A. The effect of fecal microbiota transplantation on psychiatric symptoms among patients with inflammatory bowel disease: an experimental study. Actas Esp Psiquiatr. 2020;48:1–7.
155. Gough E, Shaikh H, Manges AR. Systematic review of intestinal microbiota transplantation (fecal bacteriotherapy) for recurrent Clostridium difficile infection. Clin Infect Dis. 2011;53:994–1002.
156. Smits LP, Bouter KE, de Vos WM, Borody TJ, Nieuwdorp M. Therapeutic potential of fecal microbiota transplantation. Gastroenterology. 2013;145:946–53.
157. De Leon LM, Watson JB, Kelly CR. Transient flare of ulcerative colitis after fecal microbiota transplantation for recurrent Clostridium difficile infection. Clin Gastroenterol Hepatol. 2013;11:1036–8.
158. Brandt LJ, Aroniadis OC. An overview of fecal microbiota transplantation: techniques, indications, and outcomes. Gastrointest Endosc. 2013;78:240–9.
159. Meng X, Zhou HY, Shen HH. Microbe-metabolite-host axis, two-way action in the pathogenesis and treatment of human autoimmunity. Autoimmun Rev. 2019;18:455–75.
160. Rook GA, Raison CL, Lowry CA. Can we vaccinate against depression? Drug Discov Today. 2012;17:451–8.
161. Garay RP. Vaccinating against depression or anxiety: is it plausible? Expert Opin Biol Ther. 2017;17:525–8.
162. Lombard M, Pastoret PP, Moulin AM. A brief history of vaccines and vaccination. Rev Sci Tech. 2007;26:29–48.
163. Maes M, Kubera M, Leunis JC, Berk M. Increased IgA and IgM responses against gut commensals in chronic depression: further evidence for increased bacterial translocation or leaky gut. J Affect Disord. 2012;141:55–62.
164. Jiang H, Ling Z, Zhang Y, et al. Altered fecal microbiota composition in patients with major depressive disorder. Brain Behav Immun. 2015;48:186–94.
165. Zheng P, Zeng B, Zhou C, et al. Gut microbiome remodeling induces depressive-like behaviors through a pathway mediated by the host's metabolism. Mol Psychiatry. 2016;21:786–96.
166. Kelly JR, Borre Y, O' Brien C, et al. Transferring the blues: depression-associated gut microbiota induces neurobehavioural changes in the rat. J Psychiatr Res. 2016;82:109–18.

# Animal Inflammation-Based Models of Neuropsychiatric Disorders

5

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**Abstract**

Mounting evidence links psychiatric disorders to central and systemic inflammation. Experimental (animal) models of psychiatric disorders are important tools for translational biopsychiatry research and CNS drug discovery. Current experimental models, most typically involving rodents, continue to reveal shared fundamental pathological pathways and biomarkers underlying the pathogenetic link between brain illnesses and neuroinflammation. Recent data also show that various proinflammatory factors can alter brain neurochemistry, modulating the levels of neurohormones and neurotrophins in neurons and microglia. The role of "active" glia in releasing a wide range of proinflammatory cytokines also implicates glial cells in various psychiatric disorders. Here, we discuss recent animal inflammation-related models of psychiatric disorders, focusing on their translational perspectives and the use of some novel promising model organisms (zebrafish), to better understand the evolutionally conservative role of inflammation in neuropsychiatric conditions.

**Keywords**

Animal models · Neuroinflammation · Neurodegeneration · Rodents · Zebrafish · Model organisms

## 5.1 Introduction

Neuropsychiatric illnesses, especially affective and psychotic disorders, are the leading causes of human disability that markedly reduce the quality of life [1–5]. Mounting evidence supports the overlap of various psychiatric conditions, such as bipolar and unipolar depression, anxiety, schizophrenia, and autism, with central or peripheral inflammation [6, 7]. These two pathogenetic processes do not simply coexist, but also seem to facilitate each other, as, for example, depression promotes inflammation, whereas increased inflammation worsens depressive symptoms [7]. Chronic neuroinflammation is common in patients with psychiatric disorders [8], elevating multiple inflammatory biomarkers, such as proinflammatory cytokines and their receptors, as well as chemokines, acute-phase reactants (e.g., C-reactive protein), and adhesion molecules [9–16]. The innate immune system cells, including activated T-cells, monocytes, and neutrophils, are also hyperactive in psychiatric patients [17, 18]. While peripheral immune molecules produce pronounced behavioral alterations in both human and animal studies [19–22], remission often correlates with normalized proinflammatory biomarkers [23].

Experimental (animal) models of psychiatric disorders are important tools for translational biopsychiatry research and CNS drug discovery. Based on clear practical and ethical considerations, the most currently used experimental models of psychiatric disorders involve rodents that continue to reveal fundamental

pathological pathways and biomarkers conserved between humans and animals [24], to better translate human disease into relevant rodent phenotypes [25].

Corroborating human data, animal models show altered system of proinflammatory cytokines in various models of CNS disorders. For example, elevated levels of proinflammatory cytokines interleukins (IL) IL-6, IL-1β, and tumor necrosis factor-alpha (TNF-α) are commonly seen in rodent models of depression [26–29], strikingly paralleling clinical findings [30]. Proinflammatory factors, in turn, can also alter brain neurochemistry, including monoamine and glutamate metabolism, as well as the levels of neurohormones and neurotrophins, in neurons and microglia [6, 31–33] (Fig. 5.1). The role of "reactive" microglia (M1 phenotype) in releasing proinflammatory cytokines IL-1β, TNF-α, and IL-6 further implicates inflammation in various psychiatric disorders [6, 35–38]. Here, we discuss inflammation-related animal models of psychiatric models, focusing on their translational perspectives and novel organisms to better understand the evolutionally conservative link between inflammation and neuropsychiatric conditions.

## 5.2 Animal Models of Inflammation and CNS Disorders

Rodent experimental models are widely used in both inflammation and CNS research. Inflammation-related rodent models of psychiatric disorders often involve injecting proinflammatory substances and/or genetically manipulating various inflammation-associated genes. For instance, administering proinflammatory cytokine IL-1β to the rat anterior hypothalamus induces the release of norepinephrine, dopamine, and serotonin [39], whereas nonsteroidal anti-inflammatory drugs (NSAIDs) block murine depressive-like behavior enhanced by lipopolysaccharide (LPS) [40]. Systemic injections of LPS also induce neuroinflammation and provoke characteristic "sickness behavior" in rodents, tyopically manifesting as motor retardation (hypoactivity), decreased food and water consumption, social withdrawal, and prolonged sleep, collectively resembling signs of clinical depression [41]. During systematic inflammation, IL-1β, IL-6, TNF-α, and other proinflammatory cytokines reach various brain areas that lack a fully developed blood–brain barrier, and may also impact the hypothalamic–pituitary–adrenal (HPA) axis to trigger CNS pathogenesis.

Prenatally administered LPS or polyinosinic acid–polycytidylic acid (poly I:C) in rodents can provoke depression-like phenotype in offspring [42], whereas stress, depressive-like behavior, and aberrant sociality correlate with elevated levels of peripheral proinflammatory cytokines [43, 44]. In turn, cytokines can trigger microglial activation, elevating neuroinflammation in brain regions and thereby damaging neuronal circuits [45]. For example, following chronic stress in depression-prone mice, a neuroinflammation biomarker indoleamine-2,3-dioxygenase is increased in the raphe nuclei, whereas elevated TNF-α levels are seen in the prefrontal cortex [46].

Since neuroinflammation often leads to affective disorders clinically [47], inflammation-based rodent models of anxiety- and depression-like states become

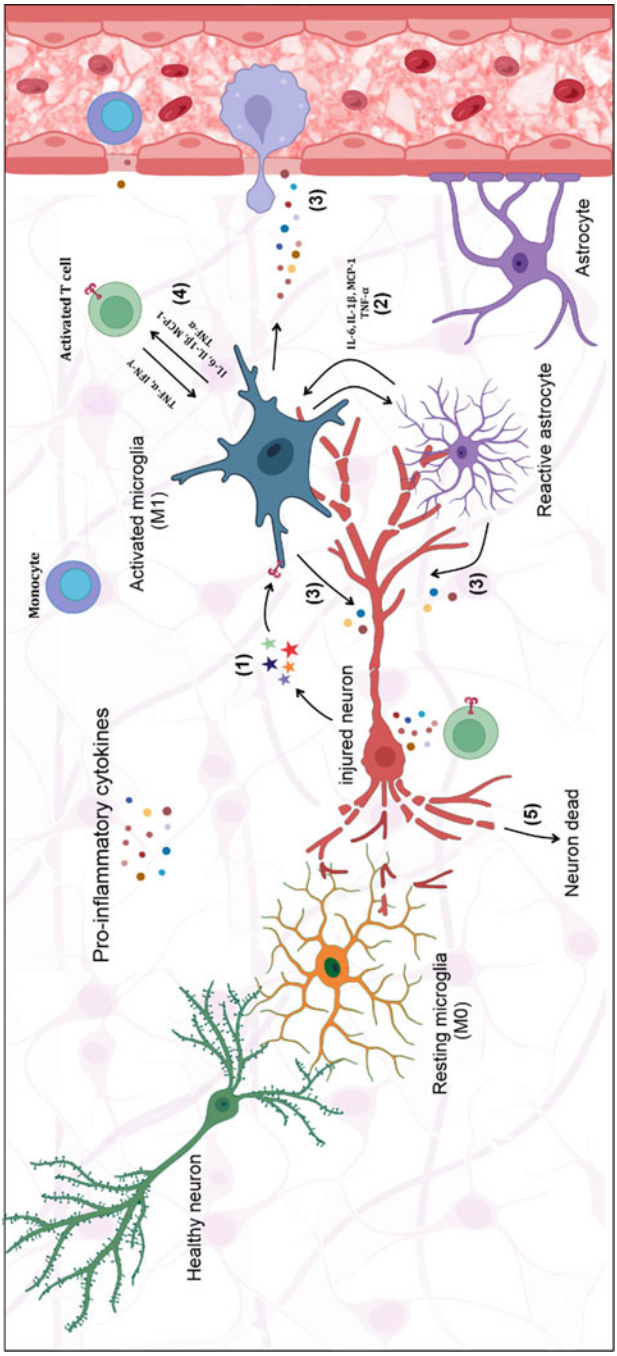


**Fig. 5.1** Neuronal death following inflammatory stimuli (according to [34]). Briefly, an injured neuron releases neural injury-derived factors (e.g., pathogen-associated molecules, (**1**)) that promote a crosstalk between neurons and other brain cells (e.g., microglia and astrocytes, (**2**)). Activated microglia and reactive astrocytes release proinflammatory cytokines (e.g., interleukins [IL] IL6, IL1β, tumor necrosis factor-alpha [TNF-α], or interferon-gamma (IFN-γ, colored balls)) which promote neuroinflammation, modulate the blood–brain barrier permeability, and attract blood cells, especially lymphocytes and monocytes (**3**). Additionally, peripheral immune cells (e.g., $CD4^+$ T cells) can also become activated, and may contribute to the inflammatory process in the brain (**4**). These proinflammatory environments and/or the lack of efficient protective mechanisms further increase inflammation, collectively triggering neuronal death (**5**)

important. For example, LPS injection provokes both inflammation and anxiety-like behavior in mice [47], as does a high-calorie carbohydrate diet [48] that also promotes inflammation and obesity. Chronic mild stress in rats not only activates microglial cells and hippocampal neuroinflammation, but evokes anxiety- and depression-like behavior as well [49].

Alzheimer's disease (AD) is another severely debilitating CNS disorder that is pathogenetically related to neuroinflammation. In rodents, a proinflammatory agent LPS promotes cognitive impairments [50] which may be relevant to modeling neurodegeneration during AD pathogenesis. Nicotinamide adenine dinucleotide (NAD+) is involved in AD and increases proinflammatory biomarkers in genetically modified APP/PS1 mice with beta-amyloid pathology [51].

Attention deficit hyperactivity disorder (ADHD) is a highly prevalent psychiatric illness characterized by inattention, impulsivity, and hyperactivity [52]. Fetal alcohol syndrome and ADHD have several common symptoms, and rat prenatal exposure to ethanol can be used as a model of ADHD [53], in line with the fact that prenatal alcohol exposure caused clinical ADHD as well [54]. Likewise, prenatally exposed animals frequently exhibit attention deficit [55, 56], impulsivity, and hyperactivity [57]—the traits that can easily be assessed in rodents in the open field, radial arm maze, or the Morris water maze test [58–60]. An opioid-like alkaloid papaverine ameliorates ADHD symptoms in rats by modulating inflammatory processes, increasing brain levels of an anti-inflammatory cytokine IL-10 and neurotrophin BDNF, as well as by lowering proinflammatory cytokines IL-6 and TNF-α [61].

Consistent with likely evolutionarily conserved pathophysiological mechanisms, neuroinflammation has particular behavioral consequences—the so-called sickness behavior that can be observed both clinically and in animals under systemic and neuroinflammatory conditions [62]. Human sickness, including systemic behavioral inhibition, loss of appetite, anhedonia, fatigue, hyperalgesia, anxiety, and neurocognitive symptoms, contributes to depression-related behavioral phenotype which is directly linked to CNS inflammation [63]. In rodents, experimentally induced sickness behavior follows the same patterns as in humans [64]. For example, IL-1β provokes depression-like behavior, HPA activation, and fever [65]. Collectively, this suggests that rodent sickness behavior is regulated by cytokines within evolutionary conserved neuroimmune mechanisms that are shared between humans and animals.

## 5.3 Zebrafish Models

A small freshwater teleost fish, the zebrafish (*Danio rerio*), has recently emerged as a promising organism in biomedicine [66–68], currently representing the second (after mice) most used laboratory animal species [69–71]. The rapidly growing use of zebrafish in neuroscience [72, 73] is supported by the simplicity of their genetic and transcription manipulations and gene editing [74, 75]; fully sequenced genome [76]; rapid and well-studied embryonic and larval development [77]; descriptive, valid, and translatable to mammals' behavioral phenotypes [78]; and the availability of medium- and high-throughput behavioral and molecular screens [73].

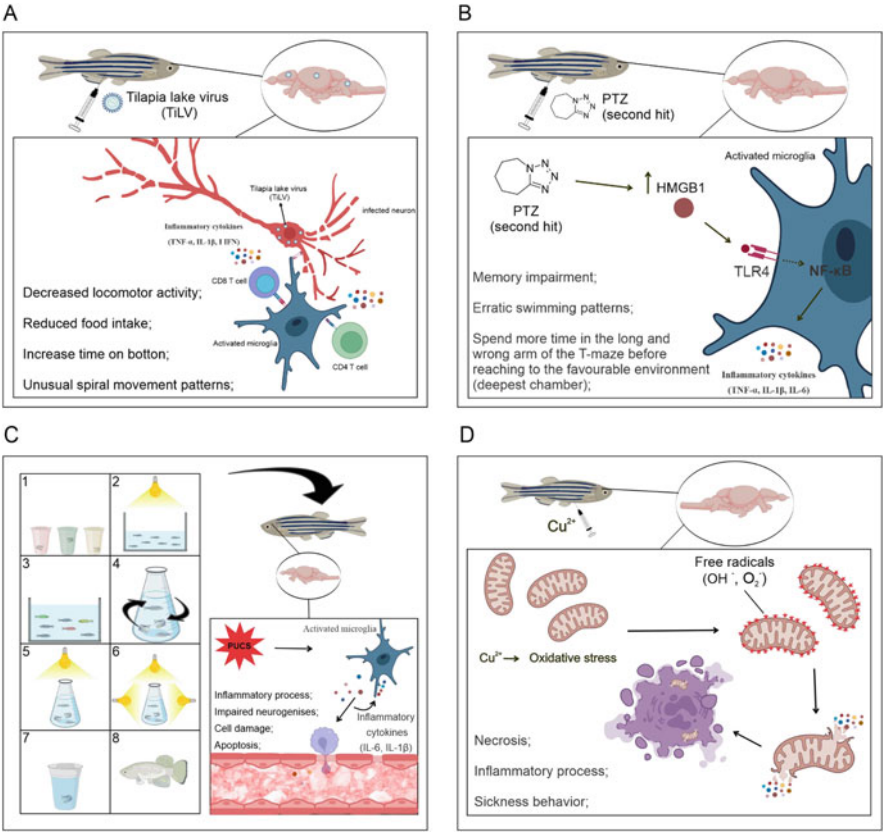


**Fig. 5.2** Selected experimental models of neuroinflammation in zebrafish. Panel (**a**) shows the tilapia lake virus (TiLV)-induced neuroinflammation model, where the injection of TiLV upregulates the expression of immune genes, activating microglia and $CD4^+$ and $CD8^+$ T cells to release proinflammatory cytokines that facilitate adult zebrafish neuroinflammation [79]. Panel (**b**) illustrates the neuroinflammation model based on a chemotoxin pentylenetetrazol (PTZ) that induces the expression of the high mobility group box 1 (HMGB1), damage-associated molecular patterns (DAMPs) that promote inflammation upon release into the extracellular space. The HMGB1 acts via the toll-like receptor 4 (TLR4) to activate the NF-κB pathways that triggers proinflammatory cytokine release [80]. Panel (**c**) shows how prolonged unpredictable chronic stress (PUCS) induces neuroinflammation in stressed zebrafish. The PUCS battery involves different types of stressors (e.g., novel object exposure, bright light with shallow water, conspecific exposure, shaking, crowding, superbright light, social isolation, exposure to a predator) that induce changes in zebrafish immune biomarkers in the brain and body, including elevated proinflammatory cytokines, which are typically produced by microglia [81]. Panel (**d**) illustrates copper ($Cu^{2+}$) injection-induced neuroinflammation in zebrafish, increasing oxidative stress in brain parenchyma and releasing free radicals that, in turn, injure cellular membranes and release proinflammatory cytokines [82]

Zebrafish are also becoming a valuable model for probing inflammation-related neuropsychiatric disorders (Fig. 5.2). For example, these fish display a wide

spectrum of aberrant CNS states, including anxiety-like, depression-like, despair-like, and anhedonia-like behaviors [83–86] that are paralleled by increased expression of proinflammatory biomarker genes of IL-1β, IL-6 [81], and COX-2 [87]. Interestingly, compared to mammals, zebrafish have generally superior neuroregenerative potential [88]. For example, unlike in mice, functional recovery of zebrafish locomotor abilities is observed in the spinal cord injury model within 4–8 weeks [89–91]. Recovery can also be seen in zebrafish models of AD [92] and traumatic brain injury [93, 94]. Thus, zebrafish can be used as a CNS disease model to study the role of neuroprotection in psychiatric illnesses, including neuroinflammation-related psychiatric disorders.

Overall, zebrafish inflammation is rather conservative and shares the same mechanisms with humans, including macrophage and neutrophil migration to the source of inflammation with further involvement of other immune cells [95]. Similarly to humans, major inflammatory mediators of zebrafish include vasoactive amines (histamine, serotonin), vasoactive peptides (substance P, bradykinin), complement components (C5aR1, C3aR1), lipid mediators (prostaglandin E2), cytokines (interleukins/ILs, interferons/INFs), chemokines (Cxcl12a, Cxcl12b, Ccl19, and Cxcl8), and proteolytic enzymes (cysteine, serine, aspartic, and metalloproteinases) [96].

Currently, zebrafish inflammation studies employ well-recognized experimental models, such as the tail fin amputation (TFA), pharmacogenic models (e.g., using LPS, leukotriene B4 [LTB4], copper, trinitrobenzene sulfonic acid [TNBS]), and genetic models (e.g., zebrafish mutants with aberrant hepatocyte growth factor activator inhibitor 1a [*hai1a*] and the cdp-diacylglycerol–inositol 3-phosphatidyltransferase [*cdipt*] genes) [97].

Neuroinflammation is a particular case of general inflammation and is based on the CNS' own immune response to specific pathologens and insults, such as brain trauma, neuroinfection, or stress [98]. The main mechanisms of neuroinflammation involves the activation of microglia and astrocytes, which initiates immune response via immune mediators, recruiting the peripheral immune cells [99]. In zebrafish, intracerebral hemorrhage (ICH) is one of the most efficient experimental models of neuroinflammation [100]. As in mammals, the functional division of microglia and brain macrophages occurs in zebrafish ICH, including the M1 (proinflammatory) and the M2 (anti-inflammatory) microglia phenotypes, respectively [101]. However, zebrafish brain is generally much more resilient to neuroinflammatory damage due to the presence of radial glia cells, which can transform into new neurons [102].

The M1 microglia starts releasing proinflammatory agents, such as IL-1β, IL-6, TNF-α, and chemokines (CXCs), that stimulate the blood–brain barrier permeability to peripheral immune cells (macrophages and T-cells), which in turn activate M2 microglia-promoted neuron progenitor cell (NPC) proliferation via anti-inflammatory factors (IGF-1, TGF-β) and specific signaling pathways, such as Stat3 and β-catenin [103].

Like mammals, zebrafish display characteristic sickness behavior (e.g., in a model of tilapia lake virus-induced neuroinflammation, Fig. 5.2), including hypolocomotion, decreased food intake, and increased anxiety [79]. Bacterial

infections induce similar behavior in zebrafish, including hypolocomotion, bottom dwelling, slow circling in the center of the experimental tank, and stereotypic intermittent stops [104]. In contrast, epilepsy-driven inflammation in zebrafish (Fig. 5.2) correlates with cognitive decline in learning and memory tasks, successfully reversed by anti-inflammatory monoclonal antibodies (e.g., anti-HMGB1 monoclonal antibody [mAb]) [105]. Hypoxia-induced neuroinflammation shifts the content of some neurotransmitters, indicating acetylcholine decrease and GABA or glutamate increase, with unaltered serotonin [106], whereas pilocarpine seizure-like activity in zebrafish elevates neuroinflammatory markers and glutamate [107].

Recent cross-species genomic studies on mammalian and zebrafish acute systemic inflammation reveal high evolutionary conservation of inflammation-related genes between these taxa, including *tlr4, il1b, il6, cxcl8a, ccl-c25y, ccl34.a, ccl35.2, ccl22, csf3, cxcl8a, cxcl11.1, il10, il1b, il6, tnfa, atf3,* and *socs1b* [108]. A well-established zebrafish systemic inflammation model, TFA, demonstrates the upregulation of prostaglandin E2 (PGE2), inducible NO synthase (iNOS), cyclooxygenase-2 (COX-2), TNF-α, IL-10, IL-6, IL-1β, and nuclear factor (NF-κB) [109]. Acute inflammation induces remarkable shifts in the expression of some key inflammatory factors in zebrafish brain, including the upregulation of the CCAAT/enhancer binding proteins (C/EBPs) of "b" and "d" isoforms, COX-2 [110].

Stress also modulates zebrafish neuroinflammation, elevating proinflammatory (IL-1β, TNF-α) and anti-inflammatory (IL-10) cytokine expression under chronic, but not acute stress [111]. Similar results were obtained with fluorescent light-induced inflammation in zebrafish brain, where in addition to IL-1β, IL-6, and TNF-α, other up-regulated proinflammatory biomarkers include INF-γ and peroxisome proliferator-activated receptor-γ (PPAR-γ) [112].

## 5.4 Conclusion

Overall, neural and systemic inflammation typically evokes evolutionarily conserved pathophysiological changes across human and animal studies. Thus, further cross-species analyses of neuroimmune mechanisms of brain disorders become critical (Fig. 5.3). Likewise, establishing specific endophenotypes of psychiatric disorders in the context of (neuro)inflammation may also open new avenues of research, eventually leading to recognizing immune-related subtypes of various brain disorders and revealing their specific neuronal circuits. In turn, this may necessitate novel models of such subtype-specific animal models and, consequently, ensuring high face, predictive, and construct validity of such inflammation-based neuropsychiatric models. Finally, widening the spectrum of animal model organisms for such translational research, including a wider use of some novel model species, such as zebrafish, is warranted.

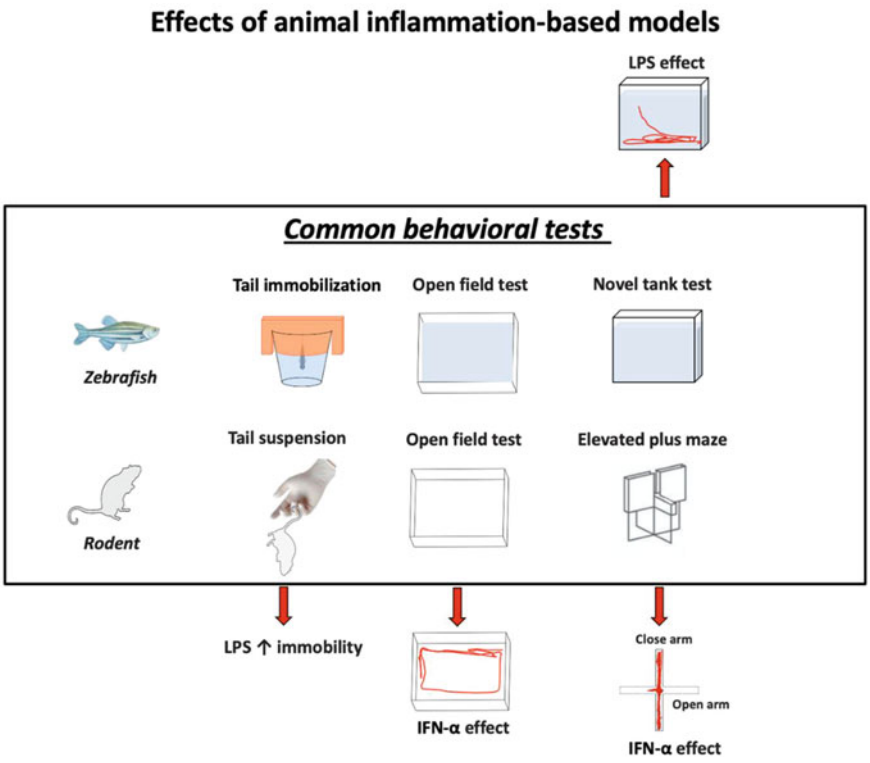


**Fig. 5.3** Selected tests to assess behavioral deficits in animal inflammation-based models. Inset: rodent and zebrafish behavioral models to measure affective (depression/anxiety-, anhedonia-, and despair-like) states [35, 113]. Other panels show examples of selected behavioral deficits seen in animal inflammation-based models. For example, mice acutely exposed to lipopolysaccharide (LPS), spend more time immobile in the tail suspension test of behavioral "despair" [114]. Mice acutely exposed to interferon (IFN-α) spend less time in the central zone and travel a shorter distance in the open arms of the elevated plus maze test, indicative of anxiogenic-like effect [115]. Likewise, zebrafish chronically exposed to LPS spend less time at the top of the tank, an common "affective" anxiety-like behavior in fish [116]

## References

1. Murray CJ, Lopez AD. Alternative projections of mortality and disability by cause 1990–2020: global burden of disease study. Lancet. 1997;349(9064):1498–504.
2. Insel TR, Charney DS. Research on major depression: strategies and priorities. JAMA. 2003;289(23):3167–8.
3. Rapaport MH, et al. Quality-of-life impairment in depressive and anxiety disorders. Am J Psychiatr. 2005;162(6):1171–8.
4. Stein MB, et al. Functional impact and health utility of anxiety disorders in primary care outpatients. Med Care. 2005;43:1164–70.

5. Walker ER, McGee RE, Druss BG. Mortality in mental disorders and global disease burden implications: a systematic review and meta-analysis. JAMA Psychiat. 2015;72(4):334–41.
6. Réus GZ, et al. The role of inflammation and microglial activation in the pathophysiology of psychiatric disorders. Neuroscience. 2015;300:141–54.
7. Bauer ME, Teixeira AL. Inflammation in psychiatric disorders: what comes first? Ann N Y Acad Sci. 2019;1437(1):57–67.
8. Najjar S, et al. Neuroinflammation and psychiatric illness. J Neuroinflammation. 2013;10(1): 1–24.
9. Modabbernia A, et al. Cytokine alterations in bipolar disorder: a meta-analysis of 30 studies. Biol Psychiatry. 2013;74(1):15–25.
10. Dargél AA, et al. C-reactive protein alterations in bipolar disorder: a meta-analysis. J Clin Psychiatry. 2015;76(2):3919.
11. Köhler CA, et al. Peripheral cytokine and chemokine alterations in depression: a meta-analysis of 82 studies. Acta Psychiatr Scand. 2017;135(5):373–87.
12. Inoshita M, et al. A significant causal association between C-reactive protein levels and schizophrenia. Sci Rep. 2016;6(1):1–8.
13. Goldsmith D, Rapaport M, Miller B. A meta-analysis of blood cytokine network alterations in psychiatric patients: comparisons between schizophrenia, bipolar disorder and depression. Mol Psychiatry. 2016;21(12):1696–709.
14. Valkanova V, Ebmeier KP, Allan CL. CRP, IL-6 and depression: a systematic review and meta-analysis of longitudinal studies. J Affect Disord. 2013;150(3):736–44.
15. Miller BJ, et al. Meta-analysis of cytokine alterations in schizophrenia: clinical status and antipsychotic effects. Biol Psychiatry. 2011;70(7):663–71.
16. Khandaker GM, et al. Inflammation and immunity in schizophrenia: implications for pathophysiology and treatment. Lancet Psychiatry. 2015;2(3):258–70.
17. do Prado CH, et al. Reduced regulatory T cells are associated with higher levels of Th1/TH17 cytokines and activated MAPK in type 1 bipolar disorder. Psychoneuroendocrinology. 2013;38(5):667–76.
18. Barbosa IG, et al. Monocyte and lymphocyte activation in bipolar disorder: a new piece in the puzzle of immune dysfunction in mood disorders. Int J Neuropsychopharmacol. 2015;18(1): pyu021.
19. Dantzer R, et al. From inflammation to sickness and depression: when the immune system subjugates the brain. Nat Rev Neurosci. 2008;9(1):46–56.
20. Harrison NA, et al. Inflammation causes mood changes through alterations in subgenual cingulate activity and mesolimbic connectivity. Biol Psychiatry. 2009;66(5):407–14.
21. Eisenberger NI, et al. Inflammation-induced anhedonia: endotoxin reduces ventral striatum responses to reward. Biol Psychiatry. 2010;68(8):748–54.
22. Haroon E, Raison CL, Miller AH. Psychoneuroimmunology meets neuropsychopharmacology: translational implications of the impact of inflammation on behavior. Neuropsychopharmacology. 2012;37(1):137–62.
23. Cattaneo A, et al. Candidate genes expression profile associated with antidepressants response in the GENDEP study: differentiating between baseline 'predictors' and longitudinal 'targets'. Neuropsychopharmacology. 2013;38(3):377–85.
24. Kalueff A, Wheaton M, Murphy D. What's wrong with my mouse model?: advances and strategies in animal modeling of anxiety and depression. Behav Brain Res. 2007;179(1):1–18.
25. Crawley JN. What's wrong with my mouse?: behavioral phenotyping of transgenic and knockout mice. Hoboken: John Wiley & Sons; 2007.
26. Wieck A, Andersen SL, Brenhouse HC. Evidence for a neuroinflammatory mechanism in delayed effects of early life adversity in rats: relationship to cortical NMDA receptor expression. Brain Behav Immun. 2013;28:218–26.
27. Mutlu O, et al. Effects of fluoxetine, tianeptine and olanzapine on unpredictable chronic mild stress-induced depression-like behavior in mice. Life Sci. 2012;91(25–26):1252–62.

28. Hanke M, et al. Beta adrenergic blockade decreases the immunomodulatory effects of social disruption stress. Brain Behav Immun. 2012;26(7):1150–9.
29. Carboni L, et al. Early-life stress and antidepressants modulate peripheral biomarkers in a gene–environment rat model of depression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2010;34(6):1037–48.
30. Yuan N, et al. Inflammation-related biomarkers in major psychiatric disorders: a cross-disorder assessment of reproducibility and specificity in 43 meta-analyses. Transl Psychiatry. 2019;9 (1):1–13.
31. Furtado M, Katzman MA. Examining the role of neuroinflammation in major depression. Psychiatry Res. 2015;229(1–2):27–36.
32. Felger JC, Lotrich FE. Inflammatory cytokines in depression: neurobiological mechanisms and therapeutic implications. Neuroscience. 2013;246:199–229.
33. Lotrich FE. Inflammatory cytokine-associated depression. Brain Res. 2015;1617:113–25.
34. de Araujo Boleti AP, et al. Neuroinflammation: an overview of neurodegenerative and metabolic diseases and of biotechnological studies. Neurochem Int. 2020;136:104714.
35. Ma L, et al. Animal inflammation-based models of depression and their application to drug discovery. Expert Opin Drug Discovery. 2017;12(10):995–1009.
36. Sellgren CM, et al. Increased synapse elimination by microglia in schizophrenia patient-derived models of synaptic pruning. Nat Neurosci. 2019;22(3):374–85.
37. Wang M, Zhang L, Gage FH. Microglia, complement and schizophrenia. Nat Neurosci. 2019;22(3):333–4.
38. Lee J-S, et al. Antidepressant-like activity of myelophil via attenuation of microglial-mediated neuroinflammation in mice undergoing unpredictable chronic mild stress. Front Pharmacol. 2019;10:683.
39. Shintani F, et al. Interleukin-1 beta augments release of norepinephrine, dopamine, and serotonin in the rat anterior hypothalamus. J Neurosci. 1993;13(8):3574–81.
40. de Paiva VN, et al. Prostaglandins mediate depressive-like behaviour induced by endotoxin in mice. Behav Brain Res. 2010;215(1):146–51.
41. Norden DM, et al. Sequential activation of microglia and astrocyte cytokine expression precedes increased Iba-1 or GFAP immunoreactivity following systemic immune challenge. Glia. 2016;64(2):300–16.
42. Enayati M, et al. Maternal infection during late pregnancy increases anxiety—and depression-like behaviors with increasing age in male offspring. Brain Res Bull. 2012;87(2–3):295–302.
43. Christian LM, et al. Depressive symptoms are associated with elevated serum proinflammatory cytokines among pregnant women. Brain Behav Immun. 2009;23(6):750–4.
44. Hodes GE, et al. Neuroimmune mechanisms of depression. Nat Neurosci. 2015;18(10): 1386–93.
45. Kempuraj D, et al. Neuroinflammation induces neurodegeneration. J Neurol Neurosurg Spine. 2016;1(1):1003.
46. Couch Y, et al. Microglial activation, increased TNF and SERT expression in the prefrontal cortex define stress-altered behaviour in mice susceptible to anhedonia. Brain Behav Immun. 2013;29:136–46.
47. Zheng ZH, et al. Neuroinflammation induces anxiety- and depressive-like behavior by modulating neuronal plasticity in the basolateral amygdala. Brain Behav Immun. 2021;91: 505–18.
48. Gomes JAS, et al. High-refined carbohydrate diet consumption induces neuroinflammation and anxiety-like behavior in mice. J Nutr Biochem. 2020;77:108317.
49. Wang YL, et al. Microglial activation mediates chronic mild stress-induced depressive- and anxiety-like behavior in adult rats. J Neuroinflammation. 2018;15(1):21.
50. Zakaria R, et al. Lipopolysaccharide-induced memory impairment in rats: a model of Alzheimer's disease. Physiol Res. 2017;66(4):553–65.

51. Hou Y, et al. NAD(+) supplementation reduces neuroinflammation and cell senescence in a transgenic mouse model of Alzheimer's disease via cGAS-STING. Proc Natl Acad Sci U S A. 2021;118(37):e2011226118.
52. Edition F. Diagnostic and statistical manual of mental disorders. Am Psychiatric Assoc. 2013;21:591–643.
53. Atalar EG, Uzbay T, Karakas S. Modeling symptoms of attention-deficit hyperactivity disorder in a rat model of fetal alcohol syndrome. Alcohol Alcohol. 2016;51(6):684–90.
54. Rojas-Mayorquin AE, Padilla-Velarde E, Ortuno-Sahagun D. Prenatal alcohol exposure in rodents as a promising model for the study of ADHD molecular basis. Front Neurosci. 2016;10:565.
55. Leth-Steensen C, Elbaz ZK, Douglas VI. Mean response times, variability, and skew in the responding of ADHD children: a response time distributional approach. Acta Psychol. 2000;104(2):167–90.
56. Hausknecht KA, et al. Prenatal alcohol exposure causes attention deficits in male rats. Behav Neurosci. 2005;119(1):302.
57. Gilbertson RJ, Barron S. Neonatal ethanol and nicotine exposure causes locomotor activity changes in preweanling animals. Pharmacol Biochem Behav. 2005;81(1):54–64.
58. Girard T, et al. Early postnatal ethanol exposure has long-term effects on the performance of male rats in a delayed matching-to-place task in the Morris water maze. Alcohol Clin Exp Res. 2000;24(3):300–6.
59. Reyes E, Wolfe J, Savage DD. The effects of prenatal alcohol exposure on radial arm maze performance in adult rats. Physiol Behav. 1989;46(1):45–8.
60. Nagahara AH, Handa RJ. Fetal alcohol exposure produces delay-dependent memory deficits in juvenile and adult rats. Alcohol Clin Exp Res. 1997;21(4):710–5.
61. Sharma N, et al. Papaverine ameliorates prenatal alcohol-induced experimental attention deficit hyperactivity disorder by regulating neuronal function, inflammation, and oxidative stress. Int J Dev Neurosci. 2021;81(1):71–81.
62. Kelley KW, Kent S. The legacy of sickness behaviors. Front Psychiatry. 2020;11:607269.
63. Maes M, et al. Depression and sickness behavior are Janus-faced responses to shared inflammatory pathways. BMC Med. 2012;10(1):66.
64. Lasselin J, et al. Comparison of bacterial lipopolysaccharide-induced sickness behavior in rodents and humans: relevance for symptoms of anxiety and depression. Neurosci Biobehav Rev. 2020;115:15–24.
65. Anforth HR, et al. Biological activity and brain actions of recombinant rat interleukin-1alpha and interleukin-1beta. Eur Cytokine Netw. 1998;9(3):279–88.
66. Meshalkina DA, et al. Adult zebrafish in CNS disease modeling: a tank that's half-full, not half-empty, and still filling. Lab Anim. 2017;46(10):378–87.
67. Cofer ZC, Matthews RP. Zebrafish models of biliary atresia and other infantile cholestatic diseases. Curr Pathobiol Rep. 2014;2(2):75–83.
68. Gong Z, et al. The zebrafish model for liver carcinogenesis. In: Molecular genetics of liver neoplasia. Cham: Springer; 2010. p. 197–218.
69. Aleström P, Winther-Larsen HC. Zebrafish offer aquaculture research their services. In: Genomics in aquaculture. London: Elsevier; 2016. p. 165–94.
70. Geisler R, et al. Archiving of zebrafish lines can reduce animal experiments in biomedical research. EMBO Rep. 2017;18(1):1–2.
71. Hudson-Shore M. Statistics of scientific procedures on living animals Great Britain 2015—highlighting an ongoing upward trend in animal use and missed opportunities for reduction. Altern Lab Anim. 2016;44(6):569–80.
72. Stewart AM, et al. Molecular psychiatry of zebrafish. Mol Psychiatry. 2015;20(1):2.
73. Stewart AM, et al. Zebrafish models for translational neuroscience research: from tank to bedside. Trends Neurosci. 2014;37(5):264–78.
74. Gerlai R. Using zebrafish to unravel the genetics of complex brain disorders. Curr Top Behav Neurosci. 2011;12:3–24.

75. Le Bras A. Enhancing gene editing in zebrafish. Lab Animal. 2019;48:234.
76. Howe K, et al. The zebrafish reference genome sequence and its relationship to the human genome. Nature. 2013;496(7446):498–503.
77. Grunwald DJ, Eisen JS. Headwaters of the zebrafish—emergence of a new model vertebrate. Nat Rev Genet. 2002;3(9):717–24.
78. Kalueff AV, et al. Towards a comprehensive catalog of zebrafish behavior 1.0 and beyond. Zebrafish. 2013;10(1):70–86.
79. Mojzesz M, et al. Tilapia Lake virus-induced neuroinflammation in zebrafish: microglia activation and sickness behavior. Front Immunol. 2021;12:760882.
80. Paudel YN, Othman I, Shaikh MF. Anti-high mobility group box-1 monoclonal antibody attenuates seizure-induced cognitive decline by suppressing neuroinflammation in an adult zebrafish model. Front Pharmacol. 2020;11:613009.
81. Song C, et al. Modeling consequences of prolonged strong unpredictable stress in zebrafish: complex effects on behavior and physiology. Prog Neuro-Psychopharmacol Biol Psychiatry. 2018;81:384–94.
82. Pereira TC, Campos MM, Bogo MR. Copper toxicology, oxidative stress and inflammation using zebrafish as experimental model. J Appl Toxicol. 2016;36(7):876–85.
83. Demin KA, et al. Understanding complex dynamics of behavioral, neurochemical and transcriptomic changes induced by prolonged chronic unpredictable stress in zebrafish. Sci Rep. 2020;10(1):1–20.
84. Yang L, et al. Delayed behavioral and genomic responses to acute combined stress in zebrafish, potentially relevant to PTSD and other stress-related disorders: focus on neuroglia, neuroinflammation, apoptosis and epigenetic modulation. Behav Brain Res. 2020;389:112644.
85. Demin KA, et al. Understanding neurobehavioral effects of acute and chronic stress in zebrafish. Stress. 2021;24(1):1–18.
86. Demin KA, et al. The zebrafish tail immobilization (ZTI) test as a new tool to assess stress-related behavior and a potential screen for drugs affecting despair-like states. J Neurosci Methods. 2020;337:108637.
87. Marcon M, et al. Prevention of unpredictable chronic stress-related phenomena in zebrafish exposed to bromazepam, fluoxetine and nortriptyline. Psychopharmacology. 2016;233(21):3815–24.
88. Zabegalov KN, et al. Decoding the role of zebrafish neuroglia in CNS disease modeling. Brain Res Bull. 2021;166:44–53.
89. Becker CG, et al. L1.1 is involved in spinal cord regeneration in adult zebrafish. J Neurosci. 2004;24(36):7837–42.
90. Dias DO, Göritz C. Fibrotic scarring following lesions to the central nervous system. Matrix Biol. 2018;68:561–70.
91. Reimer MM, et al. Motor neuron regeneration in adult zebrafish. J Neurosci. 2008;28(34):8510–6.
92. Saleem S, Kannan RR. Zebrafish: an emerging real-time model system to study Alzheimer's disease and neurospecific drug discovery. Cell Death Discov. 2018;4(1):1–13.
93. Kishimoto N, Shimizu K, Sawamoto K. Neuronal regeneration in a zebrafish model of adult brain injury. Dis Model Mech. 2012;5(2):200–9.
94. Cacialli P, Palladino A, Lucini C. Role of brain-derived neurotrophic factor during the regenerative response after traumatic brain injury in adult zebrafish. Neural Regen Res. 2018;13(6):941.
95. Novoa B, Figueras A. Zebrafish: model for the study of inflammation and the innate immune response to infectious diseases. Adv Exp Med Biol. 2012;946:253–75.
96. Zanandrea R, Bonan CD, Campos MM. Zebrafish as a model for inflammation and drug discovery. Drug Discov Today. 2020;25(12):2201–11.
97. Xie Y, Meijer AH, Schaaf MJM. Modeling inflammation in zebrafish for the development of anti-inflammatory drugs. Front Cell Dev Biol. 2021;8:620984.

98. DiSabato DJ, Quan N, Godbout JP. Neuroinflammation: the devil is in the details. J Neurochem. 2016;139(Suppl 2(Suppl 2)):136–53.
99. Chiu C-C, et al. Neuroinflammation in animal models of traumatic brain injury. J Neurosci Methods. 2016;272:38–49.
100. Au-Crilly S, et al. Using zebrafish larvae to study the pathological consequences of hemorrhagic stroke. JoVE. 2019;148:e59716.
101. Crilly S, et al. Using zebrafish larval models to study brain injury, locomotor and neuroinflammatory outcomes following intracerebral haemorrhage. F1000Res. 2018;7:1617.
102. Kyritsis N, et al. Acute inflammation initiates the regenerative response in the adult zebrafish brain. Science. 2012;338(6112):1353–6.
103. Kanagaraj P, et al. Microglia stimulate zebrafish brain repair via a specific inflammatory cascade. bioRxiv. 2020; p. 2020.10.08.330662.
104. Lee S-B, et al. Analysis of zebrafish (Danio rerio) behavior in response to bacterial infection using a self-organizing map. BMC Vet Res. 2015;11(1):269.
105. Paudel YN, Othman I, Shaikh MF. Anti-high mobility group Box-1 monoclonal antibody attenuates seizure-induced cognitive decline by suppressing neuroinflammation in an adult zebrafish model. Front Pharmacol. 2021;11:613009.
106. Lee Y, et al. Hypoxia-induced neuroinflammation and learning-memory impairments in adult zebrafish are suppressed by glucosamine. Mol Neurobiol. 2018;55(11):8738–53.
107. Paudel YN, et al. Pilocarpine induced behavioral and biochemical alterations in chronic seizure-like condition in adult zebrafish. Int J Mol Sci. 2020;21(7):2492.
108. Forn-Cuní G, et al. Conserved gene regulation during acute inflammation between zebrafish and mammals. Sci Rep. 2017;7(1):41905.
109. Hong J-M, et al. Anti-Inflammatory effects of Antarctic Lichen *Umbilicaria antarctica* methanol extract in lipopolysaccharide-stimulated RAW 264.7 macrophage cells and zebrafish model. Biomed Res Int. 2021;2021:8812090.
110. Lefebvre KA, et al. Gene expression profiles in zebrafish brain after acute exposure to domoic acid at symptomatic and asymptomatic doses. Toxicol Sci. 2009;107(1):65–77.
111. Kirsten K, et al. Acute and chronic stress differently alter the expression of cytokine and neuronal markers genes in zebrafish brain. Stress. 2021;24(1):107–12.
112. Boswell M, et al. Deconvoluting wavelengths leading to fluorescent light induced inflammation and cellular stress in Zebrafish (Danio rerio). Sci Rep. 2020;10(1):3321.
113. de Abreu MS, et al. Towards modeling anhedonia and its treatment in zebrafish. Int J Neuropsychopharmacol. 2021;25(4):293–306.
114. Dunn AJ, Swiergiel AH. Effects of interleukin-1 and endotoxin in the forced swim and tail suspension tests in mice. Pharmacol Biochem Behav. 2005;81(3):688–93.
115. Zeng J, et al. Interferon-α exacerbates neuropsychiatric phenotypes in lupus-prone mice. Arthritis Res Ther. 2019;21(1):205.
116. Demin KA, et al. Modulation of behavioral and neurochemical responses of adult zebrafish by fluoxetine, eicosapentaenoic acid and lipopolysaccharide in the prolonged chronic unpredictable stress model. Sci Rep. 2021;11(1):14289.

# Early Life Stress, Neuroinflammation, and Psychiatric Illness of Adulthood

6

Sang Ho Shin and Yong-Ku Kim

**Abstract**

Stress exposure during early stages of life elevates the risk of developing psychopathologies and psychiatric illness in later life. The brain and immune system are not completely developed by birth and therefore continue develop after birth; this post birth development is influenced by several psychosocial factors; hence, early life stress (ELS) exposure can alter brain structural development and function. A growing number of experimental animal and observational human studies have investigated the link between ELS exposure and increased risk of psychopathology through alternations in the immune system, by evaluating inflammation biomarkers. Recent studies, including brain imaging, have also shed light on the mechanisms by which both the innate and adaptive immune systems interact with neural circuits and neurotransmitters, which affect psychopathology. Herein, we discuss the link between the experience of stress in early life and lifelong alterations in the immune system, which subsequently lead to the development of various psychiatric illnesses.

**Keywords**

Early life stress · Psychopathology · Psychiatric illness · Inflammation · Cytokines · Neuroinflammation · Immune system

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## 6.1 Introduction

Both positive and negative experiences in early life can have profound effects on mammalian brain development. In particular, early life stress (ELS) is associated with increased risk of both mental and physical health deterioration throughout life [1–3]. Approximately 12% of youth, from infancy to age 18 years, experiences ELS to such an extent that they will have mental and physical health discrepancies throughout life [4, 5]. These individuals who account for almost half of all mental disorders [6] experience approximately 44% increase in strokes and heart attacks [7] and elevated risk of death by age 50 years [8, 9].

Generally, two main criteria, namely, the developmental age range of early life and the characteristics of stressful events, should be considered when defining ELS. Previous studies have considered an upper age limit between 12 and 18 years as early life. Regarding the stress criterion, various models have proposed that stress is experienced when an individual faces a threatening situation for which adequate coping skills are not available. Disruption to physiological homeostasis also induces stress responses. A certain event occurring within a defined developmental term may be classified as ELS. Thus, ELS can be defined as an injury, potential of injury, or threat of injury generally caused by a child's caregiver [10]. This definition includes different stressful experiences, such as the death of a caregiver, neglect, bullying, emotional maltreatment, and physical and sexual abuse. However, emotional and physical abuse and neglect are the most common patterns of harm reported [11].

The most major forms of ELS in individuals are neglect (physical or emotional), abuse (physical, emotional, or sexual), and parental loss (death, or separation). The number of abused children reached approximately 520,000 in the United Kingdom in 2011, which has since increased [12]. According to a large-scale epidemiological study on adverse childhood events, approximately 65% of individuals in the United States have been exposed to at least one event, while 12.5% have been exposed to as many as four [13]. Adults reporting ≥4 ELS are 4.6 times more likely to experience depression and 12.2 times more likely to attempt suicide compared to those with no ELS exposure. Gilbert et al. found that children self-reporting physical and emotional abuse is estimated at up to 29% and 33%, respectively, in Eastern Europe [14].

Approximately 8% of males and 20% of females experience childhood sexual abuse, with the highest prevalence in Africa (34.4%), followed by Asia, America (10.1%), and Europe (9.2%) [15]. Physical abuse, the intentional use of physical force that harms the child's development, survival, or dignity, is estimated at 17.6% [16]. Meanwhile, psychological abuse, the failure to give children enough supportive environment, may also be more than physical and sexual abuse but is more difficult to estimate [17]. Neglect, the failure of a caregiver to provide for a child's basic needs, is the most major ELS affecting 78.5% of children in the general population. Other forms of ELS include natural disasters, physical diseases, surgeries, accidents, and events such as terrorism or war. Less salient experiences with significant distress on children include poverty, unstable families, poor parental care, and dysfunctional relationships between parent and children. ELS can be often

complex, with different forms simultaneously coexisting and can happen as chronic or ongoing stress.

ELSs are associated with an elevated risk of noncommunicable diseases in adulthood [18–21] and premature mortality [22], possibly mediated by a dysfunctional immune system, particularly chronic low-grade inflammation [23–25]. The dysregulated immune responses could be prone to developmental programming attributed to ELS exposure that only trigger an excessive stress response at onset, but also influencing long-term stress responses, leading to chronic low-grade inflammation [26, 27]. The immune system responds to foreign invaders [28], and both human and animal researches have shown that ELS can cause persistent inflammation, which could develop psychiatric problems through effects on brain development and response to stressors [29].

The brain and immune system are not fully developed at birth, yet have minimal functions in newborns that enable adjustment to limited and expected stimuli. The ongoing maturity of the immune system throughout infant and childhood indicates that environmental effects and stimulation during childhood can seriously affect the immune system. Therefore, the brain and immune system experience during early postnatal development progressively increases their repertoire to maximize adaptation to stimuli specific to the individual's own environment [1, 30, 31]. Notably, ELS gives rise to various aberrations in brain circuitry, cognitive function, and general health [32–34] and the immune system may also play a unifying role in the pathophysiology of these multifactorial diseases related to ELS. Herein, we provide an overview of the current evidence connecting ELS to elevated inflammation and subsequent risk of psychiatric disorders.

## 6.2 Early Life Stress and Inflammation

### 6.2.1 Experimental Animal Studies

The first report for the effect of ELS on the immune system came to light from experimental animal models more than half a century ago. Mouse handled before the process of weaning exhibited a decreased rate of development in transplanted tumor [35] and elevated serum antibody titer in response to the bacterial protein flagellin [36]. These results attracted interest in the area of developmental psychoneuroimmunology [37–39], which facilitated subsequent studies on the association between ELS and immune functioning in later life in rats and nonhuman primates [40, 41].

Experimental animal models have expanded our understanding of the relationship between ELS and immune system abnormalities and allowed for invasive procedures to investigate immune function since components in the immune system can be targeted with drugs during and after ELS to determine the adverse health outcomes. ELS in rats has been manipulated through various experimental models with heterogeneous effects on parental caregiving behavior, such as nursery rearing, maternal separation (MS), maternal deprivation, neonatal handling, and dexamethasone exposure. Measures of immune function range from pro-inflammatory

cytokines and chemokines in the plasma and antigen-specific immune response to pro-inflammatory gene expression in the brain and gut microbiota.

MS has been commonly used in animal models of ELS. In nonhuman primates, MS increased macrophage activity [42] and upregulated long-term pro-inflammatory gene transcription in monocytes [43]. In rats, MS elevated core temperature [40] and pro-inflammatory cytokines in the plasma [44, 45]. These findings demonstrated the association between MS and inflammatory processes in later life.

Using a mouse model, a previous study has reported that MS results in a loss of prefrontal cortex (PFC) interneurons [46], underlying a supposed mechanism of schizophrenia associated with inflammatory and excitotoxic damage [47]. In an animal model undergoing repeated MS (RMS), elevated hippocampal interleukin-1β (IL-1β) mRNA levels approximately 20 times that of the control have been reported [48]. One study also showed elevated NF-α expression in the PFC of animals sacrificed on the day of their final MS episode [49], while another reported that MS animals sacrificed immediately after their final episode had higher hypothalamic tumor necrosis factor-α (TNF-α) than those with a single episode of MS sacrificed simultaneously [48]. In animals sacrificed on the final day of MS, elevated interleukin-10 (IL-10) expression was identified in the PFC and small intestine but not the hippocampus or serum [49, 50].

In pubs, MS during brain development is associated with reduced lipopolysaccharide-binding protein expression in the hippocampus [51] and decreased microglial cell numbers in the midbrain [52]. In contrast, early MS in adult animals increases synaptic levels of pro-inflammatory cytokine interleukin-1 (IL-1) receptor [53], elevates the number and motility of cortical microglial processes [54], and exacerbates microglial activation [55]. Moreover, mice experiencing MS have a higher elevation in body core temperature after a second MS, increased cytokine expression followed by viral infection [56], and greater cortical microglial activation following exposure to chronic food-restriction stress [55]. Although the peripheral response to ELS may not be mostly activated or suppressed, ELS-linked early immune programming seems to sensitize later pro-inflammatory processes and result in higher risk to depression and anxiety in adulthood [57]. Increased heart rate and inflammatory responses to a physiological stressor [58], as well as elevated TNF-α and interferon-γ (IFN-γ) and corticosterone levels, and anxiety-like behavior [59] in maternally deprived rodents have been shown.

ELS studies have recently reported overall increases in activation and number of microglial cells in various brain regions. Microglial cells sensitized in early life could show a dysregulated response and morphological changes in later life [60]. ELS may therefore convert a neuroprotective state to a pro-inflammatory state in microglia [48]. Moreover, microglial activation and increases could facilitate brain maturation [61]. Ex vivo studies on the early MS-induced damage on microglia have reported an overall elevation in the proportion of cells with an activated morphology in the hippocampus [48, 62, 63] and medulla [64]. Furthermore, using captured microglial cells in vivo, one study showed that somatosensory stimulation in adulthood caused a significantly higher increase in microglial motility

in MS mice compared to controls which could also affect microglia–synapse interactions and neuronal function [54].

Psychosocial stressors other than immune stimuli can also provoke a microglial response that may induce different responses to threats [65]. Gong et al. reported that 1 day of brief social isolation at postnatal day (P)14 elevated microglial density in the hippocampus, presumably by facilitating increases of these cells [66]. Four days following isolation (P14–17), the number of cells was restored to normal levels. In contrast, a week of brief social isolation (P14–21) in adulthood decreased microglial cell number in the dentate gyrus of the hippocampus. Concordantly, social defeat in adolescent mice triggers early increase of ionized calcium-binding adapter molecule (IBA-1) in the hippocampus and a following decrease in microglial cells and IBA-1 expression in adulthood [67, 68]. Furthermore, a milder social defeat paradigm adopted during the adolescent stage elevated microglia number, IBA-1 expression, and the size of soma in the ventral tegmental area of pups [69]. Adult mice experiencing repeated social defeat show significant elevation in neutrophils and $CD11b^{+}LyC6^{high}$ monocytes in the spleen and circulation [70, 71]. Splenic dendritic cells from mice experiencing repeated social defeat have shown greater surface expression of major histocompatibility complex class I, CD80, and CD44, suggesting an activated state [72]. Exposure to repeated social defeat in mice and low socioeconomic status in humans can also lead to a relative expansion of a transcriptional protein associated with immature pro-inflammatory monocytes in peripheral blood mononuclear cells [73]. Therefore, various types of social stressors in early life independently impact the development of the immune system, although the dysfunctional relationship between mother and infant may negatively affect health outcome.

Recent studies on the gut microbiota found that MS in rodent and nonhuman primate models also has transient and long-term effects on gut microbiota [59, 74]. ELS-induced changes of the microbiome in murine models continue during adulthood [75–77] and are linked to anxiety-like behaviors and activation of systems involved in stress [78]. Rats exposed to stress show inflammation, altered gastrointestinal function and leaky gut, and disturbances of immune activity [78]. Moreover, gut microbiota and dysregulated inflammation in rats or mice exposed to stress can regulate the metabolism of tryptophan to kynurenine or 5-hydroxytryptamine (5-HT) [79–81]. Inflammatory cytokines such as IFN-γ and interleukin-6 (IL-6) enhance indoleamine-2,3-dioxygenase (IDO) production, which subsequently metabolizes tryptophan to kynurenine, increasing kynurenine production and decreasing 5-HT levels [80]. Elevated kynurenine/tryptophan ratios have been recognized in rats with depression-like behavior, together with elevated pro-inflammatory cytokines and altered gut microbiota [81]. Furthermore, a study on *Flexibacter* and *Prevotella*, in connection with colitis, revealed that they were more abundant in the gut of MS rats [82], and concordantly, Wong et al. showed that caspase-1 inhibition, an inflammasome factor, restored stress-induced gut microbiota alterations [83].

## 6.3 Early Life Stress and Inflammation

### 6.3.1 Observational Human Studies

ELS can affect the immune system at the time of exposure [57, 84] and alter its normal developmental trajectory [85]. The exaggerated effects of ELS on the immune system are long term, resulting in chronic low-grade inflammation throughout life [86]. A large population-based study of almost 12,000 participants observed an association between increased white blood cell counts and ELS [87].

A meta-analysis demonstrated that adults with ELS have higher levels of C-reactive protein (CRP) and the major pro-inflammatory cytokines IL-6 and TNF-α as compared to adults without ELS [88]. Another meta-analysis reported a significant correlation between ELS and inflammatory markers, with effect sizes being greatest for TNF-α, followed by IL-6 and CRP. A recent meta-analysis showed a relationship between ELS and IL-1β, IL-6, TNF-α, and CRP, but not interleukin-2 (IL-2), interleukin-4 (IL-4), interleukin-8 (IL-8), IL-10, or fibrinogen [89]. Moreover, Baumeister et al. reported that various types of ELS exposure differentially resulted in alterations of inflammatory markers [88]. Notably, physical and sexual abuse were associated with significantly elevated TNF-α and IL-6, but not CRP, which is mainly associated with parental absence during the early developmental period. Although the underlying pathophysiology remains nebulous, factors such as situation and duration of stress exposure may interact with individual stress types to regulate immune response.

Furthermore, a meta-analysis of 27 studies conducted by Kuhlman et al. evaluated the relation between ELS and inflammation in those under 18 years of age [90] and found small associations between ELS and inflammation that were statistically significant only for CRP. Meanwhile, other longitudinal studies have reported an association between ELS and elevated inflammatory markers in early adulthood [91, 92], thereby providing evidence for the relationships between ELS and increased peripheral CRP levels, particularly profound in those who develop subsequent depression in adult life [93, 94]. Danese et al. reported elevated inflammation levels in abused children who had depression at the age of 12 years compared to normal controls [95]. Increased CRP levels were also identified in 10-year-old children with recent onset of maltreatment and a genotype predisposing to elevated CRP levels [93]. Exposure to stressful events before the age of 8 years is associated with increased CRP at 10 and 15 years old [25]. Peripheral low-grade inflammation may describe the relationships between early-life stress and various physical or mental health outcomes [96–98]. To the best of our knowledge, there has been one study investigating the relationship between ELS and inflammation in preschool-aged children (3–5 years) which showed increased IL-1β levels in connection with ELS [99]. In healthy community samples, ELS has been related to elevated IL-6, in response to the Trier Social Stress Test [100] and IL-1β, interleukin-12 (IL-12), and TNF-α levels [101]. Furthermore, recent findings suggest that repeated exposure to ELS is connected with significant increases in soluble urokinase plasminogen activator receptor (suPAR) in young adulthood [102] and mid-adulthood

[92]. During activated immune and pro-inflammatory states, suPAR is discharged into the systemic bloodstream by cleavage of the membrane-bound urokinase receptor (uPAR) [103, 104] expressed on endothelial cells and immune cells [105]. While CRP and IL-6 are influenced by acute fluctuations in inflammation, such as during infections [106, 107], suPAR reflects a person's overall immune activity level and is predicted to be involved in low-grade chronic inflammation, tissue and organ damage [108, 109], development and progression of disease, adverse clinical outcomes, and mortality [110, 111] and thus is an effective and additional measure of persistent inflammatory response. Concordantly, suPAR is associated with ELS [92, 102] and adult stressful life events [112], whereas IL-6 and CRP are not persistently related to these kinds of stressors.

Granulocyte function, evaluated by ex vivo killing of *Staphylococcus aureus*, is decreased by 20% in children with divorced parents [113], which is in line with the findings of elevated vulnerability to *Staphylococcus aureus* in children exposed to acute or chronic family stress [114]. Moreover, reduced natural killer (NK) cell activity was related to stressful events in adolescents with depression or conduct disorder [115] and adult females with breast cancer [116]. However, increased NK cell activity has been identified in children whose parents showed more chronic stress, which is also associated with greater rates of febrile diseases in childhood, while this was not associated with NK cell activity [117]. Furthermore, Wyman et al. studied a younger population undergoing immune assessment [117] and observed an elevation in NK cell activity [118]. Meanwhile, Ayaydin et al., in a relatively small number of participants, also showed no significant difference in NK cell activity between control and sexually abused adolescents [119]. Sexual or physical abuse is associated with lower salivary IgA levels in young females, even though adult sexual victimization appeared to mediate this relationship [120].

Evidence of the relationship between ELS and increased reactive oxygen species (ROS) production, oxidative stress, and mitochondrial activity, which are associated with pro-inflammatory cytokine from different immune cells, has been reported [121]. Dysfunctional cellular immunity caused by repeated viral infections and reactivation of viruses elevates inflammatory markers including IL-6 and CRP [122]. Also, individuals exposed to ELS showed increased immune activation with higher CD25 expression, major histocompatibility complex, class II, DR (HLA-DR), or implicating CD8 T cells [123]. Moreover, adolescents exposed to ELS have decreased NK and NK T cells and increased circulating and senescent T cells with the activation markers CD3+/CD69+ and CD2+/CD4+/CD25+ [124, 125].

Positron emission tomography (PET) imaging of the mitochondrial translocator protein (TSPO) can provide insights on the microglial activation in the human brain. One study found that after peripheral lipopolysaccharide (LPS) injection, TSPO expression uniformly increased across the brain [126]. These findings allow for deeper assessment of neuroinflammatory markers to investigate microglial activation during brain injury and neurodegeneration. Interestingly, only one study has investigated microglial activation using TSPO-PET in individuals exposed to ELS [71, 127]; therefore, further investigations on the relationship between ELS and microglial activation are warranted.

Also, in line with findings from animal studies, results from human studies suggest that females may have higher sensitivity to stressful events in provoking a pro-inflammatory response than males [128, 129]. Moreover, evidence from an environmental risk study indicates that the levels of inflammation were already increased in children exposed to ELS who developed depression at the age of 12 years as compared to controls [95], while sex differences may influence the susceptibility to cause a pro-inflammatory state post ELS exposure [130]. CRP concentrations in 18-year-old females were significantly correlated with childhood victimization, yet no such correlation was observed in their male counterparts. According to the study by Entringer et al., the relationship between CRP levels and maltreatment was significantly mediated by child sex and were higher in the maltreated girls compared to the control group which was stable over the 2-year follow-up period, whereas no relationship between maltreatment and CRP levels was shown, suggesting that following ELS exposure at a very young age in girls, the effect of maltreatment may immediately emerge in an inflammation process and exacerbate over time.

In summary, peripheral inflammation caused by ELS can influence the brain and change neural activity through various routes, such as humorally via active transport of cytokines stimulated by the release of second messenger or cellular routes involving macrophage-like cells residing in circumventricular organs. Microglia can be activated by peripheral inflammation that enter the brain across the blood-brain barrier (BBB) with different routes [131]. Subsequently, microglia cells affect cell proliferation and survival in the brain based on their inflammatory state [126]. Microglial cells can undergo several alterations [132] such as pro-inflammatory cytokine production and expression of cell surface antigens related to oxidative stress in the brain. Peripheral LPS injection, used for immune challenge in primates, can increase TSPO expression uniformly across the brain. Recently, alterations in the gut microbiome have been reported in adults with ELS-induced PTSD [133]. Children exposed to ELS have also been reported in a study to exhibit gut microbiome alterations, with gut bacteria levels associated with PFC activation in an emotional face experiment [134]. Generally, the association between ELS-linked gut dysbiosis and inflammation is likely bidirectional [135, 136].

## 6.4 Inflammation and Psychiatric Illness

### 6.4.1 Experimental Animal Studies

Animal models are beneficial to investigate the pathophysiology due to their flexibility in randomly assigning animals to different rearing environments and allow for directly investigating the brain and immune system using techniques thought to be too invasive in humans. Inflammation can increase animals' responses to rewarding stimuli with reinforcers such as food or electrical stimulation [137]. Initial findings linking the immune function and psychiatric etiology, particularly mood disorders, originated from studies involving humans and animals with acute infection showing

stereotypical behaviors as featured by anhedonia, anorexia, and reduced grooming [138]. In line with the hypothesis of this "sickness behavior" with evolutionarily inflammatory origins, gene knockout models in rodents have been strongly beneficial in emphasizing the causal relationship of inflammatory cytokines (including IL-1β and TNF-α) in developing social withdrawal, sickness behaviors, and anhedonia [137]. Also, the development of sickness behavior led by a pro-inflammatory state is attenuated by treatment with IL-10 and aggravated in mice that are IL-10-deficient [137]. An elevation in cytokine serum levels may correspondingly elevate oxidative stress and reduce availability of serotonin and other neurotransmitters, along with activities of the hypothalamic-pituitary-adrenal (HPA) axis in the brain [137, 139]. Acute induction of pro-inflammatory agents, such as LPS or typhoid vaccination, can trigger transient and similar symptoms [140]. Rodents exposed to MS show dysfunction in PFC-mediated behaviors including social interactions [141], learned helplessness [142], and cognitive function [46] in adolescence and elevated peripheral inflammatory cytokines IL-β and IL-6 [45]. Findings in rodents have suggested that this immune-to-brain traffic can control the cortico-amygdala circuitry involved in threat processing and is connected with enhanced anxiety-like behaviors [143–145]. Pigs with MS show sickness-like behavior that is buffered with anti-inflammatory treatment [146, 147], indicating that pro-inflammatory processes can influence early responses to ELS. Social withdrawal, lethargy, and anhedonia related to exposure to pro-inflammatory agents may be part of the organism's evolutionary effort to use all its resources for fighting foreign invaders and overcoming diseases [148]. Giovanoli et al. have investigated if an anti-inflammatory medication with minocycline in early life during peripubertal adversity exposure could prevent the following occurrence of adult behavioral problems [149]. Notably, rats deficient in the inflammasome NLRP3 showed improvement in both pro-inflammatory state and cognitive function and reduced both systemic inflammation and functional decline during aging [150].

## 6.5 Inflammation and Psychiatric Illness

### 6.5.1 Observational an Experimental Human Studies

Experimental findings suggest that inflammation can decrease neural activity to reward circuit, as shown by studies that induced inflammation with low-dose bacterial stimuli [151] or investigating the effects of immune-activating treatments on neural reward circuit [152]. Induction of pro-inflammatory states in humans produces a clinical response similar to major depression [153]. Patients with some types of cancer or hepatitis C treated with interferon-α (IFN-α) also develop depressive symptoms within weeks [140, 154]. Additional experimental evidence related to the inflammation as the pathophysiology of mood disorders comes from the antidepressant effects of anti-inflammatory medications. These experimental human studies proposed that inflammation can modulate neural circuit activity linked to rewards

independently in different processes that may lead inflammation in those exposed to ELS.

Recently, a meta-analysis showed that cytokine inhibitors and nonsteroidal anti-inflammatory drugs can have small to moderate antidepressant effects [155]. Moreover, patients administered minocycline exhibited a greater decrease in negative symptoms in two clinical trials comparing minocycline versus placebo [156, 157]. Pro-inflammatory cytokines may also reduce executive control-related processes associated with PFC in the brain where it is linked to decision making, executive control, and regulation of reward and threat-related predisposition [158, 159]. Cytokine increase may alternate microglia in the cortex, thereby causing structural and functional changes, which increases the risk of mental illness [160]. Concordantly, alterations in microglial activation have been observed in several psychiatric disorders including schizophrenia [161], depression [162], and anxiety [163].

Microglia plays a major role in the adaptive immune response in the central nervous system (CNS) that can modulate neuronal function not only during inflammation but also in synaptic pruning [164] and plasticity during development [165, 166]. A recent TSPO-PET study showed elevated microglial activity in patients with schizophrenia and persons who are even at ultrahigh risk of psychosis. Moreover, increased microglial activity was positively associated with greater symptom severity in the high-risk population [167], suggesting a relationship between neuroinflammation and psychotic symptoms.

In line with these findings, human observational studies over the past 30 years have emphasized the role of the immune function in the pathophysiology of several psychiatric disorders, including schizophrenia, depression, bipolar affective disorder [168, 169], obsessive–compulsive disorder [170, 171], and posttraumatic stress disorder (PTSD), along with an increase of suicidal attempt [172]. A meta-analysis controlling the effect of antipsychotics in schizophrenia showed persistently increased levels of several immune proteins released from macrophages, such as IL-12, TNF-α, and IFN-α [97]. Interestingly, cell cultures from patients with schizophrenia also produce greater levels of circulating IL-1 and IL-8, thereby confirming the role of immunity-related pathophysiology in schizophrenia. Studies on obsessive–compulsive disorder reported polymorphisms in the TNF-α gene [173] and elevation in plasma TNF-α cytokine levels [174–176]; based on the individual, cytokine gene polymorphisms may manifest differently [173]. Other prospective studies also showed that elevated IL-6 and CRP were significantly associated with depressive symptoms later in life. Longitudinal studies have found that increase inflammatory levels in patients with depression likely result from a bidirectional relationship between inflammation and depression over time [177]. A meta-analysis of clinical studies found that patients with depression show a slight elevation in several inflammatory biomarkers [178]. Concordantly, longitudinal associations between inflammation and subsequent psychopathology were shown in participants with psychosis [179], depression [171], and PTSD [180, 181].

Associations between inflammation and psychopathology have been best investigated in depression [182]. Patients with depression show immune cell profiles

featured by systemic low-grade inflammation [183]. A cross-sectional meta-analysis investigated alterations in inflammation in depressed adults and characterized depression by a small increase of serum inflammatory markers [178]. Anti-inflammatory medications showed antidepressant effects in a subset of depressed patients with elevated baseline levels of inflammation [184]. Group differences between inflammation in patients with depression and controls likely attributed to the bidirectional relationship between depression and inflammation [29, 177]. Increased levels of pro-inflammatory cytokines, such as IL-6 and TNF-α, are associated with depressed mood [185–187], and decreased levels of the anti-inflammatory cytokine IL-10 have been shown in depression [188]. A meta-analysis demonstrated increased TNF-α and IL-6 in patients with depression [189]. Moreover, a study by Miller and Cole showed that the transition to depression was associated with relative increases in CRP and IL-6 levels in individuals exposed to ELS, indicating that ELS can potentially enhance a phenotype wherein depression and inflammation occurred simultaneously [190].

Concordantly, patients with bipolar disorder also have increased levels of inflammation [191], TLR-mediated intracellular signaling [45], and toll-like receptors (TLRs) in peripheral monocytes and lymphocytes. Moreover, elevated NLRP3 levels were found in the frontal cortex of patients with bipolar disorder, which is associated with elevated levels of IL-6, IL-1, TNF-α, and IL-10 [192]. Meta-analyses of clinical studies found that patients with bipolar disorder have small to moderate elevation of both pro-inflammatory cytokines [193] and CRP [194] levels compared to controls. An elevated inflammation state can predict poor treatment prognosis in bipolar disorder [195]. These relationships can reflect the negative outcomes in individuals exposed to ELS [29]. Systemic inflammation in patients with bipolar disorder can be identified not only during active episodes, but also in euthymic phases [194], indicating that inflammation may be a trait marker rather than a state marker for bipolar disorder.

Although there have been limited findings from cross-sectional human studies, increased IL-6 and CRP levels is associated with psychosis [3, 196], as supported by longitudinal studies involving the general population, including the Avon Longitudinal Study of Parents and Children birth cohort. Furthermore, greater levels of pro-inflammatory cytokines in childhood are associated with an elevated risk for psychosis in adolescence and young adulthood [179, 197]. A meta-analysis controlling antipsychotics persistently showed increased TNF-α, interleukin-12 (IL-12), and IFN-γ in patients with schizophrenia [97]. Furthermore, schizophrenic patients show a moderate to large increase in pro-inflammatory cytokines [97] and CRP [198]. Initial evidence also suggests that elevated baseline inflammatory levels can be predictive of poor treatment response in first-episode psychosis [126]. Indeed, a study on both chronic psychotic disorders and first-episode psychosis showed that several inflammatory markers appear to be trait markers and showed no reduction following antipsychotic medication [97, 198].

Patients with PTSD also exhibited increased inflammation levels. A meta-analysis suggests that patients with PTSD have moderate to large elevation in several pro-inflammatory cytokines [199] after controlling the effect of comorbid depression

[199]. Genetic [181] and longitudinal [180] studies have suggested that inflammation can be a preexisting susceptibility factor for patients with PTSD exposed to ELS rather than a simple correlation of disease severity, subjective distress, or dysfunctional coping strategies after PTSD development.

## 6.6 Early Life Stress and Psychiatric Illness

### 6.6.1 Experimental Animal Studies

Animal studies of ELS on psychiatric disorders found that associations between early contexts of stress and later emotional and behavioral abnormalities are likely causal in nature [200–206]. Studies using mouse [202, 205] and nonhuman primate models [203, 204, 206] have found that ELS from MS can negatively influence the emotional and behavioral development and impair cognitive functioning, in line with the seminal studies of clinical observations by Spitz [207] and Bowlby [208] on the effects of MS on psychiatric disorders. Indeed, animals exposed to ELS show behavioral despair and learned helplessness [200], dysfunctional fear conditioning [200], and avoidant behaviors. Surprisingly, sensitization in guinea pigs was first identified when two, 3-h separations at a 24-h interval increased the number of 1-min intervals that guinea pig pups spent showing a passive, depressive-like response on the second day of separation [40]. Although the effects of induced ELS can be different based on the protocol used and the animals' gender and age, the findings of these experimental studies strongly indicate a causative role of ELS in psychopathology in the late stage.

## 6.7 Early Life Stress and Psychiatric Illness

### 6.7.1 Observational Human Studies

Individuals exposed to ELS are 1.3–3.1 times more likely to result in lifetime major depressive disorder or dysthymia, based on the frequency, type and severity, and stressful events [209–211]. Although exposure to ELS can increase the risk of many psychiatric disorders, the relationship between ELS and various types of psychiatric etiology have not been clarified [6, 212–215]. One study showed that ELS predicts several psychiatric disorders, including schizophrenia, depression, bipolar disorder, and PTSD [216–218]. ELS from childhood neglect has also been related to later changes in reward function in individuals [219]. Activation of the nucleus accumbens [220] and other basal ganglia regions [221] associated with the reward system decreases in teenagers exposed to ELS. Therefore, individuals exposed to ELS have an increased lifelong risk of major depression including an early-onset and elevated comorbidity [213, 222]. Individuals with present depression and a history of ELS are also more likely to show high levels of high-sensitivity CRP compared to controls. Notably, this association is not likely to be suggested by retrospectively

biased reports of individuals in depression at the time of ELS assessment as the evidence is persistent with those from official records and prospective evaluations of maltreatment investigated in childhood [223]. Moreover, this is also unlikely to be described by the effects confounded genetically because a higher risk of depression in individuals exposed to ELS has been identified within twin studies [209].

Also, a history of ELS is highly associated with patients with bipolar disorder and can predict an unfavorable illness course and clinical symptoms such as higher severity of manic, psychotic, and depressive symptoms, a higher suicidal attempt, higher risk of comorbid substance use disorders, anxiety disorders, elevated risk of rapid cycling, and increased occurrence of depressive and manic episodes [224]. Moreover, ELS predicts an increased number of psychotic disorders such as schizophrenia or schizoaffective disorder later in life [217] Furthermore, ELS is related to an elevated risk of PTSD [218] and is associated with more complex symptoms including dysfunctional interrelationship, dysregulated emotion, and poor self-concept [225].

## 6.8 Discussion

In this chapter, we provided an overview of the literature on early-life stress, inflammation, and psychiatric illness. This section reviews how ELS affects the psychopathology of psychiatric illness via inflammation. In the past, the brain is thought to be immune-privileged with highly controlled innate and adaptive immunity, especially inflammation in the blood-brain barrier. It has increasingly become evident that the immune-privileged property of the brain is complicated and not absolute [226]. The brain immune system is not only associated with the peripheral immune system [137] but also actively contributes to normal brain development and functioning [227]. The immune system in the brain has different cells, such as T cells and microglia, and proteins such as chemokines or cytokines that play essential roles to maintain homeostasis in the CNS resting state. Microglia monitor the surrounding extracellular space during the resting state for infection and eliminate cellular debris as well as maintain neurogenesis and inactive or dysfunctional synaptic structures. Conversely, during a pro-inflammatory state, microglia produce inflammatory cytokines and other molecules and clean up triggering foreign invaders through phagocytosis. T cells originating from the lymphoid hematopoietic cell scan and detect signals cascaded from brain into the CSF during the resting state. Meanwhile, during the pro-inflammatory state, T cells release cytokines (e.g., IL-4) that stimulate astrocytes to lead the production of brain-derived neurotrophic factor (BDNF) and control inflammatory activity in parenchymal and meningeal myeloid cells such as microglia and induce a protective immune response that may be associated with aggravated results for brain function. Moreover, cytokines accumulating in the microglia and T cells at the resting states play a critical role in hippocampus-linked learning and memory processes, putatively via supporting long-term potentiation, whereas cytokines during the pro-inflammatory state enhance neuroinflammation and decrease monoaminergic transmission and trigger glutamate transmission and

the HPA axis–mediated neuroendocrine stress response. Furthermore, a higher level of cytokines inhibits BDNF and cholinergic transmission [137, 227]. Notably, there are various routes through which inflammatory cytokines can increase synaptic monoamine availability; these routes can play a fundamental role in the mechanism underlying the pathophysiology of psychiatric illness [228]. An increased level of IDO is also associated with cytokine-induced monoamine neurotransmitter changes by converting the metabolism of tryptophan more into the kynurenine pathway but less into the 5-hydroxyindoleacetic acid, thereby reducing serotonin production. Subsequently, the neurotoxic metabolite quinolinic acid from microglia, monocytes, and macrophages in the CNS originates from kynurenine [229, 230]. Quinolinic acid stimulates N-methyl-D-aspartate receptors for glutamate and glutamate release by astrocyte and blocks glutamate reuptake by astrocytes [231], which directly affect glutamate metabolism to ultimately increase excitotoxicity and decrease efficient neurogenesis, finally resulting in increased glutamate both inside and outside the synapse. Therefore, elevated glutamate also increases excitotoxicity and decreases the production of BDNF [232].

Concordantly, high levels of nitric oxide (NO) [233] released from microglia in the inflammatory state can promote more neuronal cytotoxicity and apoptosis [234, 235] and contribute to neuronal loss in schizophrenia and Alzheimer's diseases [236–238]. Thus, ELS sensitize microglial activation resulting in a lower threshold for a reactive state and subsequently increasing inflammatory cytokine levels and dysregulated neurotransmission, which can explain psychopathologies of psychiatric illness caused by ELS.

As mentioned above, previous studies have linked the peripheral immune system and the brain immune system; researchers have recognized that the immune-privileged property of the brain is complicated and not absolute. The humoral pathway refers to the cytokine passage through regions such as the circumventricular organs with increased permeability in the BBB and elevated binding of cytokines to saturable transport molecules on the BBB. The neural pathway [137] refers to the binding of peripheral cytokines to peripheral afferent nerve fibers, such as the vagus nerve, which subsequently triggers ascending catecholaminergic fibers in the CNS and/or brings back cytokine signals in the central part [139]. The signal transduction pathway refers to the triggering by peripheral cytokines from cell surface receptors on endothelial cells and astrocytes in the brain that maintain the BBB, subsequently stimulating more cytokine production by these cells. The transmembrane pathway refers to the active transport of cytokines (TNF-α, IL-6, IL-1) through saturable carrier proteins to enter the BBB. Finally, the cellular pathway refers to the trafficking of activated immune cells, typically monocytes, to the brain vasculature and parenchyma. Through these pathways, peripheral inflammation can trigger neuroinflammation in the brain [137, 239]. For example, peripheral induction of LPS in rodents increases the production of pro-inflammatory cytokines [240] and microglia activation and inhibited adult neurogenesis in the brain [241].

Chronic stress in early life causes repeated and prolonged HPA overactivation, which can subsequently cause less compensation in reduced signaling through epigenetic alterations in the glucocorticoid receptor [242] and promote resistance

to the function of cortisol to control the inflammatory state. Experimental human studies have found that traumatic experiences during childhood are associated with allele-specific DNA demethylation related to glucocorticoid response elements (FKBP5 gene), which is related to the subsequently reduced sensitivity of peripheral blood immune cells to the inhibitory function of glucocorticoids on LPS-induced production of IL-6 in vitro [243]. A longitudinal study also showed that adolescents exposed to harsh familial treatment showed decreased sensitivity of glucocorticoid over time and elevated ex vivo cytokine responses to LPS administration [244].

Furthermore, the alteration of colonization and composition of the gut microbiota might be influenced by ELS, which could affect immune development as well as brain development via inflammatory signal transmission through metabolic alterations or the vagus nerve [135]. Experimental animals findings also showed that MS during the first year of life causes a significant reduction in fecal lactobacilli [74] with long-term alterations on the composition of the microbiota in the gut being apparent in later life [59].

Interestingly, recent meta-analytical findings in animal models showed that ELS is linked to a small increase in the risk of obesity [19], as individuals with ELS may be less sensitive to reward and hence may be involved in more dysfunctional appetitive behaviors, such as eating fast foods or more high-calorie food items. Also, given that ELS can potentiate HPA axis activation and related unpleasant feelings, individuals with ELS eat more to decrease HPA axis activation. Elevated pro-inflammatory cytokines by adipocytes can trigger a systemic inflammatory state in individuals with obesity [245]. Moreover, individuals with ELS may have dysfunction in hormonal pathways regulating thermogenesis and lipolysis including the leptin pathway or the HPA axis [19].

Previous studies have also reported that individuals exposed to ELS are at increased risk of sleep disturbances [246, 247], which showed stronger relationship for participants with more severe maltreatment exposures [246] regardless of concurrent PTSD or depressive disorders [248] Furthermore, MS in rodents can disrupt sleep architecture and decrease total sleep; meanwhile, in humans, MS can induce sleep deprivation and loss, which elevates the expression and levels of pro-inflammatory cytokines [249, 250].

In line with the biological and evolutional aspects of the bidirectional associations between the brain and immune system, the critical targets primary related to inflammation in the brain include those brain regions associated with both motivation and motor activity such as arousal, anxiety, and alarm. In other words, the main neurocircuits affected by inflammation involve the reward and anxiety circuits. Dopamine as a core neurotransmitter plays a critical role in the reward circuit and inflammatory cytokines have been demonstrated to reduce the production of dopamine in the basal ganglia, which is involved in decreased motivation and activation of the reward circuit in the ventral striatum [151, 152, 251]. Accumulated imaging studies such as PET, functional magnetic resonance imaging (fMRI), and magnetic resonance spectroscopy (MRS) have shown decreases in reward activation in the striatum, showing strong reproducibility and validity of the cytokine-induced alterations of the brain in nondepressed individuals peripherally administered LPS,

typhoid vaccination, or IFN-α [151, 152, 252–254]. Notably, fMRI studies also found that inflammation-mediated reduction of positive reward activation is related to elevated sensitivity to negative stimuli and decreased activations in the substantia nigra in the basal ganglia [254, 255]. According to studies on positive valence systems, peripheral administration of typhoid vaccine and LPS decreases responses to reward in the ventral striatum [151, 152]. Inflammatory cytokines in dopaminergic pathways also induce a state of reduced motivation. Moreover, elevated inflammation is related to elevated responses to anxiety and threat neurocircuitry, involving the amygdala, dACC, and insula [155, 256, 257]. Notably, the dACC and amygdala are regions with elevated responses in patients with depression, anxiety, and neuroticism [258]. Thus, elevated oral IL-6 expression is strongly associated with increased response of the amygdala to social evaluation stressor, with subjects showing the highest IL-6 responses to stress, indicating greatest functional connectivity within threat circuitry, which involves the dorsomedial PFC and amygdala [259]. Similarly, elevated concentrations of oral IL-6 and soluble TNF receptor 2 in response to an induced social anxiety condition, such as a public speaking, are strongly associated with the activation of the dACC to a social rejection task [257]. Indeed, these findings are related to the trafficking of monocytes to the amygdala led by social defeat stress in rodents [145]. Considering negative valence systems, typhoid vaccine decreases the relationship between the sACC and the amygdala and elevates the activation in the sACC while processing emotional faces [158]; peripheral administration of LPS also enhances activation in the amygdala in conditions of socially threatening stimuli [260]. Subsequently, alterations in reward and threat processing are critical potential mediators led by the effect of systemic inflammation on behavioral responses.

As immune stimulation also profoundly affects the perinatal brain development processes involved in cognitive function, some experimental animal studies showed that infection and systemic inflammation during prenatal or neonatal periods impair learning, memory, and attention [261–264]. Meanwhile, observational human studies found a relationship between prenatal exposure to infection and elevated risk of schizophrenia [265, 266]. Elevated levels of the systemic inflammatory marker IL-6 during childhood are significantly related to an elevated risk of causing psychosis and depression in young adult [179].

Accumulated experimental and observational studies in animals and humans have suggested bidirectional relationships between psychiatric illness and inflammation in peripheral and neuroinflammation over time [177], indicating that susceptibilities associated with emotional and behavioral symptoms and dysfunctional perception of distress could elevate inflammatory responses and sensitization over time or vice versa. Thus, the severities and frequencies of stressful events could be affected by an individual's susceptibilities, such as personality traits or attachment style, and their environmental risk factors, both of which are critical risk factors for ELS [267].

## References


1. Danese A, McEwen BS. Adverse childhood experiences, allostasis, allostatic load, and age-related disease. Physiol Behav. 2012;106(1):29–39.
2. Ehlert U. Enduring psychobiological effects of childhood adversity. Psychoneuroendocrinology. 2013;38(9):1850–7.
3. Miller GE, Chen E, Parker KJ. Psychological stress in childhood and susceptibility to the chronic diseases of aging: moving toward a model of behavioral and biological mechanisms. Psychol Bull. 2011;137(6):959–97.
4. Felitti VJ, Anda RF, Nordenberg D, Williamson DF, Spitz AM, Edwards V, et al. Relationship of childhood abuse and household dysfunction to many of the leading causes of death in adults. The adverse childhood experiences (ACE) study. Am J Prev Med. 1998;14(4):245–58.
5. Kalmakis KA, Chandler GE. Health consequences of adverse childhood experiences: a systematic review. J Am Assoc Nurse Pract. 2015;27(8):457–65.
6. Kessler RC, McLaughlin KA, Green JG, Gruber MJ, Sampson NA, Zaslavsky AM, et al. Childhood adversities and adult psychopathology in the WHO world mental health surveys. Br J Psychiatry. 2010;197(5):378–85.
7. Korkeila J, Vahtera J, Nabi H, Kivimaki M, Korkeila K, Sumanen M, et al. Childhood adversities, adulthood life events and depression. J Affect Disord. 2010;127(1–3):130–8.
8. Chen E, Turiano NA, Mroczek DK, Miller GE. Association of reports of childhood abuse and all-cause mortality rates in women. JAMA Psychiat. 2016;73(9):920–7.
9. Kelly-Irving M, Lepage B, Dedieu D, Bartley M, Blane D, Grosclaude P, et al. Adverse childhood experiences and premature all-cause mortality. Eur J Epidemiol. 2013;28(9):721–34.
10. Sideli L, Mule A, La Barbera D, Murray RM. Do child abuse and maltreatment increase risk of schizophrenia? Psychiatry Investig. 2012;9(2):87–99.
11. Scherrer JF, Waterman BM, Heath AC, Bucholz KK, True WR, Jacob T. Are substance use, abuse and dependence associated with study participation? Predictors of offspring nonparticipation in a twin-family study. J Stud Alcohol. 2004;65(1):140–4.
12. Radford L, Corral S, Bradley C, Fisher H, Bassett C, Howat N, Collishaw S. Child abuse and neglect in the UK today. London: NSPCC; 2011.
13. Middlebrooks J, Auedage N. The effects of childhood stress on health across the lifespan. Atlanta, GA: CDC; 2008.
14. Gilbert R, Widom CS, Browne K, Fergusson D, Webb E, Janson S. Burden and consequences of child maltreatment in high-income countries. Lancet. 2009;373(9657):68–81.
15. Verdolini N, Attademo L, Agius M, Ferranti L, Moretti P, Quartesan R. Traumatic events in childhood and their association with psychiatric illness in the adult. Psychiatr Danub. 2015;27(Suppl 1):S60–70.
16. Dubowitz H, Pitts SC, Black MM. Measurement of three major subtypes of child neglect. Child Maltreat. 2004;9(4):344–56.
17. Holmes WC, Slap GB. Sexual abuse of boys: definition, prevalence, correlates, sequelae, and management. JAMA. 1998;280(21):1855–62.
18. Bennouna-Greene M, Bennouna-Greene V, Berna F, Defranoux L. History of abuse and neglect in patients with schizophrenia who have a history of violence. Child Abuse Negl. 2011;35(5):329–32.
19. Danese A, Tan M. Childhood maltreatment and obesity: systematic review and meta-analysis. Mol Psychiatry. 2014;19(5):544–54.
20. Hughes K, Bellis MA, Hardcastle KA, Sethi D, Butchart A, Mikton C, et al. The effect of multiple adverse childhood experiences on health: a systematic review and meta-analysis. Lancet Public Health. 2017;2(8):e356–e66.
21. Tsehay M, Necho M, Mekonnen W. The role of adverse childhood experience on depression symptom, prevalence, and severity among school going adolescents. Depress Res Treat. 2020;2020:5951792.

22. Bellis MA, Hughes K, Leckenby N, Hardcastle KA, Perkins C, Lowey H. Measuring mortality and the burden of adult disease associated with adverse childhood experiences in England: a national survey. J Public Health (Oxf). 2015;37(3):445–54.
23. Flouri E, Francesconi M, Midouhas E, Lewis G. Prenatal and childhood adverse life events, inflammation and depressive symptoms across adolescence. J Affect Disord. 2020;260:577–82.
24. O'Connor TG, Willoughby MT, Moynihan JA, Messing S, Vallejo Sefair A, Carnahan J, et al. Early childhood risk exposures and inflammation in early adolescence. Brain Behav Immun. 2020;86:22–9.
25. Slopen N, Kubzansky LD, McLaughlin KA, Koenen KC. Childhood adversity and inflammatory processes in youth: a prospective study. Psychoneuroendocrinology. 2013;38(2):188–200.
26. Bucci M, Marques SS, Oh D, Harris NB. Toxic stress in children and adolescents. Adv Pediatr Infect Dis. 2016;63(1):403–28.
27. Franke HA. Toxic stress: effects, prevention and treatment. Children (Basel). 2014;1(3):390–402.
28. Johnson SB, Riley AW, Granger DA, Riis J. The science of early life toxic stress for pediatric practice and advocacy. Pediatrics. 2013;131(2):319–27.
29. Danese A, van Harmelen AL. The hidden wounds of childhood trauma. Eur J Psychotraumatol. 2017;8(sup5):137584.
30. Bateson P, Barker D, Clutton-Brock T, Deb D, D'Udine B, Foley RA, et al. Developmental plasticity and human health. Nature. 2004;430(6998):419–21.
31. Greenough WT, Black JE, Wallace CS. Experience and brain development. Child Dev. 1987;58(3):539–59.
32. Anda RF, Brown DW, Dube SR, Bremner JD, Felitti VJ, Giles WH. Adverse childhood experiences and chronic obstructive pulmonary disease in adults. Am J Prev Med. 2008;34(5):396–403.
33. Brown DW, Anda RF, Felitti VJ, Edwards VJ, Malarcher AM, Croft JB, et al. Adverse childhood experiences are associated with the risk of lung cancer: a prospective cohort study. BMC Public Health. 2010;10:20.
34. Dube SR, Fairweather D, Pearson WS, Felitti VJ, Anda RF, Croft JB. Cumulative childhood stress and autoimmune diseases in adults. Psychosom Med. 2009;71(2):243–50.
35. Ader R, Friedman SB. Social factors affecting emotionality and resistance to disease in animals. V. Early separation from the mother and response to a transplanted tumor in the rat. Psychosom Med. 1965;27:119–22.
36. Solomon GF, Levine S, Kraft JK. Early experience and immunity. Nature. 1968;220(5169):821–2.
37. Ader R. Developmental psychoneuroimmunology. Dev Psychobiol. 1983;16(4):251–67.
38. Coe CL. Developmental psychoneuroimmunology revisited. Brain Behav Immun. 1996;10(3):185–7.
39. Danese A. Developmental psychoneuroimmunology: from bench to bedside. Brain Behav Immun. 2014;36:27–8.
40. Hennessy MB, Deak T, Schiml-Webb PA, Carlisle CW, O'Brien E. Maternal separation produces, and a second separation enhances, core temperature and passive behavioral responses in Guinea pig pups. Physiol Behav. 2010;100(4):305–10.
41. Shanks N, Lightman SL. The maternal-neonatal neuro-immune interface: are there long-term implications for inflammatory or stress-related disease? J Clin Invest. 2001;108(11):1567–73.
42. Coe CL, Rosenberg LT, Levine S. Prolonged effect of psychological disturbance on macrophage chemiluminescence in the squirrel monkey. Brain Behav Immun. 1988;2(2):151–60.
43. Cole SW, Conti G, Arevalo JM, Ruggiero AM, Heckman JJ, Suomi SJ. Transcriptional modulation of the developing immune system by early life social adversity. Proc Natl Acad Sci U S A. 2012;109(50):20578–83.

44. Reus GZ, Dos Santos MA, Abelaira HM, Ribeiro KF, Petronilho F, Vuolo F, et al. Imipramine reverses alterations in cytokines and BDNF levels induced by maternal deprivation in adult rats. Behav Brain Res. 2013;242:40–6.
45. Wieck A, Andersen SL, Brenhouse HC. Evidence for a neuroinflammatory mechanism in delayed effects of early life adversity in rats: relationship to cortical NMDA receptor expression. Brain Behav Immun. 2013;28:218–26.
46. Brenhouse HC, Andersen SL. Nonsteroidal anti-inflammatory treatment prevents delayed effects of early life stress in rats. Biol Psychiatry. 2011;70(5):434–40.
47. Behrens MM, Sejnowski TJ. Does schizophrenia arise from oxidative dysregulation of parvalbumin-interneurons in the developing cortex? Neuropharmacology. 2009;57(3): 193–200.
48. Roque A, Ochoa-Zarzosa A, Torner L. Maternal separation activates microglial cells and induces an inflammatory response in the hippocampus of male rat pups, independently of hypothalamic and peripheral cytokine levels. Brain Behav Immun. 2016;55:39–48.
49. Giridharan VV, Reus GZ, Selvaraj S, Scaini G, Barichello T, Quevedo J. Maternal deprivation increases microglial activation and neuroinflammatory markers in the prefrontal cortex and hippocampus of infant rats. J Psychiatr Res. 2019;115:13–20.
50. Moya-Perez A, Perez-Villalba A, Benitez-Paez A, Campillo I, Sanz Y. Bifidobacterium CECT 7765 modulates early stress-induced immune, neuroendocrine and behavioral alterations in mice. Brain Behav Immun. 2017;65:43–56.
51. Wei L, Simen A, Mane S, Kaffman A. Early life stress inhibits expression of a novel innate immune pathway in the developing hippocampus. Neuropsychopharmacology. 2012;37(2): 567–80.
52. Chocyk A, Dudys D, Przyborowska A, Majcher I, Mackowiak M, Wedzony K. Maternal separation affects the number, proliferation and apoptosis of glia cells in the substantia nigra and ventral tegmental area of juvenile rats. Neuroscience. 2011;173:1–18.
53. Viviani B, Boraso M, Valero M, Gardoni F, Marco EM, Llorente R, et al. Early maternal deprivation immunologically primes hippocampal synapses by redistributing interleukin-1 receptor type I in a sex dependent manner. Brain Behav Immun. 2014;35:135–43.
54. Takatsuru Y, Nabekura J, Ishikawa T, Kohsaka S, Koibuchi N. Early-life stress increases the motility of microglia in adulthood. J Physiol Sci. 2015;65(2):187–94.
55. Brenhouse HC, Thompson V. Maternal separation increases IBA-1 expression: a microglia activation marker in the prefrontal cortex of adolescent males following a second hit of stress (abstract). Soc Biol Psychiatry. 2015;77:52s.
56. Avitsur R, Hunzeker J, Sheridan JF. Role of early stress in the individual differences in host response to viral infection. Brain Behav Immun. 2006;20(4):339–48.
57. Hennessy MB, Deak T, Schiml-Webb PA. Early attachment-figure separation and increased risk for later depression: potential mediation by proinflammatory processes. Neurosci Biobehav Rev. 2010;34(6):782–90.
58. Loria AS, Pollock DM, Pollock JS. Early life stress sensitizes rats to angiotensin II-induced hypertension and vascular inflammation in adult life. Hypertension. 2010;55(2):494–9.
59. O'Mahony SM, Marchesi JR, Scully P, Codling C, Ceolho AM, Quigley EM, et al. Early life stress alters behavior, immunity, and microbiota in rats: implications for irritable bowel syndrome and psychiatric illnesses. Biol Psychiatry. 2009;65(3):263–7.
60. Tay TL, Sagar DJ, Grun D, Prinz M. Unique microglia recovery population revealed by single-cell RNAseq following neurodegeneration. Acta Neuropathol Commun. 2018;6(1):87.
61. Cowan CS, Callaghan BL, Kan JM, Richardson R. The lasting impact of early-life adversity on individuals and their descendants: potential mechanisms and hope for intervention. Genes Brain Behav. 2016;15(1):155–68.
62. Delpech JC, Wei L, Hao J, Yu X, Madore C, Butovsky O, et al. Early life stress perturbs the maturation of microglia in the developing hippocampus. Brain Behav Immun. 2016;57:79–93.

63. Saavedra LM, Fenton Navarro B, Torner L. Early life stress activates glial cells in the hippocampus but attenuates cytokine secretion in response to an immune challenge in rat pups. Neuroimmunomodulation. 2017;24(4–5):242–55.
64. Baldy C, Fournier S, Boisjoly-Villeneuve S, Tremblay ME, Kinkead R. The influence of sex and neonatal stress on medullary microglia in rat pups. Exp Physiol. 2018;103(9):1192–9.
65. Frank MG, Fonken LK, Watkins LR, Maier SF. Microglia: neuroimmune-sensors of stress. Semin Cell Dev Biol. 2019;94:176–85.
66. Gong Y, Tong L, Yang R, Hu W, Xu X, Wang W, et al. Dynamic changes in hippocampal microglia contribute to depressive-like behavior induced by early social isolation. Neuropharmacology. 2018;135:223–33.
67. Zhang Y, Xu H, Wang J, Ren F, Shao F, Ellenbroek B, et al. Transient upregulation of immune activity induced by adolescent social stress is involved in cognitive deficit in adult male mice and early intervention with minocycline. Behav Brain Res. 2019;374:112136.
68. Zhang Y, Xu H, Zhang F, Shao F, Ellenbroek B, Wang J, et al. Deficiencies of microglia and TNFalpha in the mPFC-mediated cognitive inflexibility induced by social stress during adolescence. Brain Behav Immun. 2019;79:256–66.
69. Lo Iacono L, Catale C, Martini A, Valzania A, Viscomi MT, Chiurchiu V, et al. From traumatic childhood to cocaine abuse: the critical function of the immune system. Biol Psychiatry. 2018;84(12):905–16.
70. Gomez-Gonzalez B, Escobar A. Prenatal stress alters microglial development and distribution in postnatal rat brain. Acta Neuropathol. 2010;119(3):303–15.
71. Rupprecht R, Papadopoulos V, Rammes G, Baghai TC, Fan J, Akula N, et al. Translocator protein (18 kDa) (TSPO) as a therapeutic target for neurological and psychiatric disorders. Nat Rev Drug Discov. 2010;9(12):971–88.
72. Zhao Q, Peng C, Wu X, Chen Y, Wang C, You Z. Maternal sleep deprivation inhibits hippocampal neurogenesis associated with inflammatory response in young offspring rats. Neurobiol Dis. 2014;68:57–65.
73. Zhao Q, Xie X, Fan Y, Zhang J, Jiang W, Wu X, et al. Phenotypic dysregulation of microglial activation in young offspring rats with maternal sleep deprivation-induced cognitive impairment. Sci Rep. 2015;5:9513.
74. Bailey MT, Coe CL. Maternal separation disrupts the integrity of the intestinal microflora in infant rhesus monkeys. Dev Psychobiol. 1999;35(2):146–55.
75. Garcia-Rodenas CL, Bergonzelli GE, Nutten S, Schumann A, Cherbut C, Turini M, et al. Nutritional approach to restore impaired intestinal barrier function and growth after neonatal stress in rats. J Pediatr Gastroenterol Nutr. 2006;43(1):16–24.
76. Jasarevic E, Howard CD, Misic AM, Beiting DP, Bale TL. Stress during pregnancy alters temporal and spatial dynamics of the maternal and offspring microbiome in a sex-specific manner. Sci Rep. 2017;7:44182.
77. Jasarevic E, Rodgers AB, Bale TL. A novel role for maternal stress and microbial transmission in early life programming and neurodevelopment. Neurobiol Stress. 2015;1:81–8.
78. Gareau MG, Silva MA, Perdue MH. Pathophysiological mechanisms of stress-induced intestinal damage. Curr Mol Med. 2008;8(4):274–81.
79. Carlessi AS, Borba LA, Zugno AI, Quevedo J, Reus GZ. Gut microbiota-brain axis in depression: the role of neuroinflammation. Eur J Neurosci. 2021;53(1):222–35.
80. Christmas DM, Potokar J, Davies SJ. A biological pathway linking inflammation and depression: activation of indoleamine 2,3-dioxygenase. Neuropsychiatr Dis Treat. 2011;7:431–9.
81. Kelly JR, Borre Y, Brien CO, Patterson E, El Aidy S, Deane J, et al. Transferring the blues: depression-associated gut microbiota induces neurobehavioural changes in the rat. J Psychiatr Res. 2016;82:109–18.
82. Pusceddu MM, El Aidy S, Crispie F, O'Sullivan O, Cotter P, Stanton C, et al. N-3 polyunsaturated fatty acids (PUFAs) reverse the impact of early-life stress on the Gut microbiota. PLoS One. 2015;10(10):e0139721.

83. Wong ML, Inserra A, Lewis MD, Mastronardi CA, Leong L, Choo J, et al. Inflammasome signaling affects anxiety-and depressive-like behavior and gut microbiome composition. Mol Psychiatry. 2016;21(6):797–805.
84. Hennessy MB, Paik KD, Caraway JD, Schiml PA, Deak T. Proinflammatory activity and the sensitization of depressive-like behavior during maternal separation. Behav Neurosci. 2011;125(3):426–33.
85. Coe CL, Lubach G, Ershler WB. Immunological consequences of maternal separation in infant primates. New Dir Child Dev. 1989;45:65–91.
86. Hertzman C, Boyce T. How experience gets under the skin to create gradients in developmental health. Annu Rev Public Health. 2010;31:329–47. 3p following 47
87. Surtees P, Wainwright N, Day N, Luben R, Brayne C, Khaw KT. Association of depression with peripheral leukocyte counts in EPIC-Norfolk—role of sex and cigarette smoking. J Psychosom Res. 2003;54(4):303–6.
88. Baumeister D, Akhtar R, Ciufolini S, Pariante CM, Mondelli V. Childhood trauma and adulthood inflammation: a meta-analysis of peripheral C-reactive protein, interleukin-6 and tumour necrosis factor-alpha. Mol Psychiatry. 2016;21(5):642–9.
89. Tursich M, Neufeld RW, Frewen PA, Harricharan S, Kibler JL, Rhind SG, et al. Association of trauma exposure with proinflammatory activity: a transdiagnostic meta-analysis. Transl Psychiatry. 2014;4:e413.
90. Kuhlman KR, Horn SR, Chiang JJ, Bower JE. Early life adversity exposure and circulating markers of inflammation in children and adolescents: a systematic review and meta-analysis. Brain Behav Immun. 2020;86:30–42.
91. Danese A, Pariante CM, Caspi A, Taylor A, Poulton R. Childhood maltreatment predicts adult inflammation in a life-course study. Proc Natl Acad Sci U S A. 2007;104(4):1319–24.
92. Rasmussen LJH, Moffitt TE, Eugen-Olsen J, Belsky DW, Danese A, Harrington H, et al. Cumulative childhood risk is associated with a new measure of chronic inflammation in adulthood. J Child Psychol Psychiatry. 2019;60(2):199–208.
93. Cicchetti D, Handley ED, Rogosch FA. Child maltreatment, inflammation, and internalizing symptoms: investigating the roles of C-reactive protein, gene variation, and neuroendocrine regulation. Dev Psychopathol. 2015;27(2):553–66.
94. Danese A, Moffitt TE, Pariante CM, Ambler A, Poulton R, Caspi A. Elevated inflammation levels in depressed adults with a history of childhood maltreatment. Arch Gen Psychiatry. 2008;65(4):409–15.
95. Danese A, Caspi A, Williams B, Ambler A, Sugden K, Mika J, et al. Biological embedding of stress through inflammation processes in childhood. Mol Psychiatry. 2011;16(3):244–6.
96. Fagundes CP, Glaser R, Kiecolt-Glaser JK. Stressful early life experiences and immune dysregulation across the lifespan. Brain Behav Immun. 2013;27(1):8–12.
97. Miller BJ, Buckley P, Seabolt W, Mellor A, Kirkpatrick B. Meta-analysis of cytokine alterations in schizophrenia: clinical status and antipsychotic effects. Biol Psychiatry. 2011;70(7):663–71.
98. Slavich GM, Irwin MR. From stress to inflammation and major depressive disorder: a social signal transduction theory of depression. Psychol Bull. 2014;140(3):774–815.
99. Tyrka AR, Parade SH, Valentine TR, Eslinger NM, Seifer R. Adversity in preschool-aged children: effects on salivary interleukin-1beta. Dev Psychopathol. 2015;27(2):567–76.
100. Carpenter LL, Gawuga CE, Tyrka AR, Lee JK, Anderson GM, Price LH. Association between plasma IL-6 response to acute stress and early-life adversity in healthy adults. Neuropsychopharmacology. 2010;35(13):2617–23.
101. Li L, Chassan RA, Bruer EH, Gower BA, Shelton RC. Childhood maltreatment increases the risk for visceral obesity. Obesity (Silver Spring). 2015;23(8):1625–32.
102. Rasmussen LJH, Moffitt TE, Arseneault L, Danese A, Eugen-Olsen J, Fisher HL, et al. Association of adverse experiences and exposure to violence in childhood and adolescence with inflammatory burden in young people. JAMA Pediatr. 2020;174(1):38–47.

103. Dekkers PE, ten Hove T, te Velde AA, van Deventer SJ, van Der Poll T. Upregulation of monocyte urokinase plasminogen activator receptor during human endotoxemia. Infect Immun. 2000;68(4):2156–60.
104. Ostrowski SR, Piironen T, Hoyer-Hansen G, Gerstoft J, Pedersen BK, Ullum H. Reduced release of intact and cleaved urokinase receptor in stimulated whole-blood cultures from human immunodeficiency virus-1-infected patients. Scand J Immunol. 2005;61(4):347–56.
105. Blasi F, Carmeliet P. uPAR: a versatile signalling orchestrator. Nat Rev Mol Cell Biol. 2002;3 (12):932.
106. Hunter CA, Jones SA. IL-6 as a keystone cytokine in health and disease. Nat Immunol. 2015;16(5):448–57.
107. Rhodes B, Furnrohr BG, Vyse TJ. C-reactive protein in rheumatology: biology and genetics. Nat Rev Rheumatol. 2011;7(5):282–9.
108. Desmedt S, Desmedt V, Delanghe JR, Speeckaert R, Speeckaert MM. The intriguing role of soluble urokinase receptor in inflammatory diseases. Crit Rev Clin Lab Sci. 2017;54(2): 117–33.
109. Lyngbaek S, Sehestedt T, Marott JL, Hansen TW, Olsen MH, Andersen O, et al. CRP and suPAR are differently related to anthropometry and subclinical organ damage. Int J Cardiol. 2013;167(3):781–5.
110. Eugen-Olsen J, Andersen O, Linneberg A, Ladelund S, Hansen TW, Langkilde A, et al. Circulating soluble urokinase plasminogen activator receptor predicts cancer, cardiovascular disease, diabetes and mortality in the general population. J Intern Med. 2010;268(3):296–308.
111. Rasmussen LJ, Ladelund S, Haupt TH, Ellekilde G, Poulsen JH, Iversen K, et al. Soluble urokinase plasminogen activator receptor (suPAR) in acute care: a strong marker of disease presence and severity, readmission and mortality. A retrospective cohort study. Emerg Med J. 2016;33(11):769–75.
112. Bourassa KJ, Rasmussen LJH, Danese A, Eugen-Olsen J, Harrington H, Houts R, et al. Linking stressful life events and chronic inflammation using suPAR (soluble urokinase plasminogen activator receptor). Brain Behav Immun. 2021;97:79–88.
113. Bartlett JA, Demetrikopoulos MK, Schleifer SJ, Keller SE. Phagocytosis and killing of Staphylococcus aureus: effects of stress and depression in children. Clin Diagn Lab Immunol. 1997;4(3):362–6.
114. Meyer RJ, Haggerty RJ. Streptococcal infections in families. Factors altering individual susceptibility. Pediatrics. 1962;29:539–49.
115. Birmaher B, Rabin BS, Garcia MR, Jain U, Whiteside TL, Williamson DE, et al. Cellular immunity in depressed, conduct disorder, and normal adolescents: role of adverse life events. J Am Acad Child Adolesc Psychiatry. 1994;33(5):671–8.
116. Witek Janusek L, Tell D, Albuquerque K, Mathews HL. Childhood adversity increases vulnerability for behavioral symptoms and immune dysregulation in women with breast cancer. Brain Behav Immun. 2013;30(Suppl):S149–62.
117. Wyman PA, Moynihan J, Eberly S, Cox C, Cross W, Jin X, et al. Association of family stress with natural killer cell activity and the frequency of illnesses in children. Arch Pediatr Adolesc Med. 2007;161(3):228–34.
118. Naliboff BD, Benton D, Solomon GF, Morley JE, Fahey JL, Bloom ET, et al. Immunological changes in young and old adults during brief laboratory stress. Psychosom Med. 1991;53(2): 121–32.
119. Ayaydin H, Abali O, Akdeniz NO, Kok BE, Gunes A, Yildirim A, et al. Immune system changes after sexual abuse in adolescents. Pediatr Int. 2016;58(2):105–12.
120. Waldron JC, Scarpa A, Kim-Spoon J, Coe CL. Adult sexual experiences as a mediator between child abuse and current secretory immunoglobulin a levels. J Interpers Violence. 2016;31(5):942–60.
121. Boeck C, Koenig AM, Schury K, Geiger ML, Karabatsiakis A, Wilker S, et al. Inflammation in adult women with a history of child maltreatment: the involvement of mitochondrial alterations and oxidative stress. Mitochondrion. 2016;30:197–207.

122. Bennett JM, Glaser R, Malarkey WB, Beversdorf DQ, Peng J, Kiecolt-Glaser JK. Inflammation and reactivation of latent herpesviruses in older adults. Brain Behav Immun. 2012;26(5):739–46.
123. Elwenspoek MMC, Sias K, Hengesch X, Schaan VK, Leenen FAD, Adams P, et al. T cell Immunosenescence after early life adversity: association with cytomegalovirus infection. Front Immunol. 2017;8:1263.
124. Elwenspoek MMC, Hengesch X, Leenen FAD, Schritz A, Sias K, Schaan VK, et al. Proinflammatory T cell status associated with early life adversity. J Immunol. 2017;199(12): 4046–55.
125. Elwenspoek MMC, Kuehn A, Muller CP, Turner JD. The effects of early life adversity on the immune system. Psychoneuroendocrinology. 2017;82:140–54.
126. Mondelli V, Vernon AC, Turkheimer F, Dazzan P, Pariante CM. Brain microglia in psychiatric disorders. Lancet Psychiatry. 2017;4(7):563–72.
127. Chen MK, Guilarte TR. Translocator protein 18 kDa (TSPO): molecular sensor of brain injury and repair. Pharmacol Ther. 2008;118(1):1–17.
128. Rampp C, Eichelkraut A, Best J, Czamara D, Rex-Haffner M, Uhr M, et al. Sex-related differential response to dexamethasone in endocrine and immune measures in depressed in-patients and healthy controls. J Psychiatr Res. 2018;98:107–15.
129. Rohleder N, Schommer NC, Hellhammer DH, Engel R, Kirschbaum C. Sex differences in glucocorticoid sensitivity of proinflammatory cytokine production after psychosocial stress. Psychosom Med. 2001;63(6):966–72.
130. Baldwin JR, Arseneault L, Caspi A, Fisher HL, Moffitt TE, Odgers CL, et al. Childhood victimization and inflammation in young adulthood: a genetically sensitive cohort study. Brain Behav Immun. 2018;67:211–7.
131. Cattaneo A, Macchi F, Plazzotta G, Veronica B, Bocchio-Chiavetto L, Riva MA, et al. Inflammation and neuronal plasticity: a link between childhood trauma and depression pathogenesis. Front Cell Neurosci. 2015;9:40.
132. Walker FR, Beynon SB, Jones KA, Zhao Z, Kongsui R, Cairns M, et al. Dynamic structural remodelling of microglia in health and disease: a review of the models, the signals and the mechanisms. Brain Behav Immun. 2014;37:1–14.
133. Hemmings SMJ, Malan-Muller S, van den Heuvel LL, Demmitt BA, Stanislawski MA, Smith DG, et al. The microbiome in posttraumatic stress disorder and trauma-exposed controls: an exploratory study. Psychosom Med. 2017;79(8):936–46.
134. Callaghan BL, Fields A, Gee DG, Gabard-Durnam L, Caldera C, Humphreys KL, et al. Mind and gut: associations between mood and gastrointestinal distress in children exposed to adversity. Dev Psychopathol. 2020;32(1):309–28.
135. Cryan JF, Dinan TG. Mind-altering microorganisms: the impact of the gut microbiota on brain and behaviour. Nat Rev Neurosci. 2012;13(10):701–12.
136. Hooper LV, Littman DR, Macpherson AJ. Interactions between the microbiota and the immune system. Science. 2012;336(6086):1268–73.
137. Dantzer R, O'Connor JC, Freund GG, Johnson RW, Kelley KW. From inflammation to sickness and depression: when the immune system subjugates the brain. Nat Rev Neurosci. 2008;9(1):46–56.
138. Hart BL. Biological basis of the behavior of sick animals. Neurosci Biobehav Rev. 1988;12 (2):123–37.
139. Miller AH, Maletic V, Raison CL. Inflammation and its discontents: the role of cytokines in the pathophysiology of major depression. Biol Psychiatry. 2009;65(9):732–41.
140. Reichenberg A, Yirmiya R, Schuld A, Kraus T, Haack M, Morag A, et al. Cytokine-associated emotional and cognitive disturbances in humans. Arch Gen Psychiatry. 2001;58(5):445–52.
141. Holland FH, Ganguly P, Potter DN, Chartoff EH, Brenhouse HC. Early life stress disrupts social behavior and prefrontal cortex parvalbumin interneurons at an earlier time-point in females than in males. Neurosci Lett. 2014;566:131–6.

142. Leussis MP, Freund N, Brenhouse HC, Thompson BS, Andersen SL. Depressive-like behavior in adolescents after maternal separation: sex differences, controllability, and GABA. Dev Neurosci. 2012;34(2–3):210–7.
143. Frank MG, Watkins LR, Maier SF. Stress- and glucocorticoid-induced priming of neuroinflammatory responses: potential mechanisms of stress-induced vulnerability to drugs of abuse. Brain Behav Immun. 2011;25(Suppl 1):S21–8.
144. Wohleb ES, Hanke ML, Corona AW, Powell ND, Stiner LM, Bailey MT, et al. ta-adrenergic receptor antagonism prevents anxiety-like behavior and microglial reactivity induced by repeated social defeat. J Neurosci. 2011;31(17):6277–88.
145. Wohleb ES, Powell ND, Godbout JP, Sheridan JF. Stress-induced recruitment of bone marrow-derived monocytes to the brain promotes anxiety-like behavior. J Neurosci. 2013;33 (34):13820–33.
146. Hennessy MB, Schiml-Webb PA, Miller EE, Maken DS, Bullinger KL, Deak T. Anti-inflammatory agents attenuate the passive responses of Guinea pig pups: evidence for stress-induced sickness behavior during maternal separation. Psychoneuroendocrinology. 2007;32 (5):508–15.
147. Perkeybile AM, Schiml-Webb PA, O'Brien E, Deak T, Hennessy MB. Anti-inflammatory influences on behavioral, but not cortisol, responses during maternal separation. Psychoneuroendocrinology. 2009;34(7):1101–8.
148. Hartung HP, Heininger K, Schafer B, Fierz W, Toyka KV. Immune mechanisms in inflammatory polyneuropathy. Ann N Y Acad Sci. 1988;540:122–61.
149. Giovanoli S, Engler H, Engler A, Richetto J, Feldon J, Riva MA, et al. Preventive effects of minocycline in a neurodevelopmental two-hit model with relevance to schizophrenia. Transl Psychiatry. 2016;6:e772.
150. Youm YH, Grant RW, McCabe LR, Albarado DC, Nguyen KY, Ravussin A, et al. Canonical Nlrp3 inflammasome links systemic low-grade inflammation to functional decline in aging. Cell Metab. 2013;18(4):519–32.
151. Eisenberger NI, Berkman ET, Inagaki TK, Rameson LT, Mashal NM, Irwin MR. Inflammation-induced anhedonia: endotoxin reduces ventral striatum responses to reward. Biol Psychiatry. 2010;68(8):748–54.
152. Capuron L, Pagnoni G, Drake DF, Woolwine BJ, Spivey JR, Crowe RJ, et al. Dopaminergic mechanisms of reduced basal ganglia responses to hedonic reward during interferon alfa administration. Arch Gen Psychiatry. 2012;69(10):1044–53.
153. Smith RS. The macrophage theory of depression. Med Hypotheses. 1991;35(4):298–306.
154. Musselman DL, Lawson DH, Gumnick JF, Manatunga AK, Penna S, Goodkin RS, et al. Paroxetine for the prevention of depression induced by high-dose interferon alfa. N Engl J Med. 2001;344(13):961–6.
155. Kohler O, Benros ME, Nordentoft M, Farkouh ME, Iyengar RL, Mors O, et al. Effect of anti-inflammatory treatment on depression, depressive symptoms, and adverse effects: a systematic review and meta-analysis of randomized clinical trials. JAMA Psychiat. 2014;71(12): 1381–91.
156. Chaudhry IB, Hallak J, Husain N, Minhas F, Stirling J, Richardson P, et al. Minocycline benefits negative symptoms in early schizophrenia: a randomised double-blind placebo-controlled clinical trial in patients on standard treatment. J Psychopharmacol. 2012;26(9): 1185–93.
157. Levkovitz Y, Mendlovich S, Riwkes S, Braw Y, Levkovitch-Verbin H, Gal G, et al. A double-blind, randomized study of minocycline for the treatment of negative and cognitive symptoms in early-phase schizophrenia. J Clin Psychiatry. 2010;71(2):138–49.
158. Harrison NA, Brydon L, Walker C, Gray MA, Steptoe A, Critchley HD. Inflammation causes mood changes through alterations in subgenual cingulate activity and mesolimbic connectivity. Biol Psychiatry. 2009;66(5):407–14.

159. Juengling FD, Ebert D, Gut O, Engelbrecht MA, Rasenack J, Nitzsche EU, et al. Prefrontal cortical hypometabolism during low-dose interferon alpha treatment. Psychopharmacology. 2000;152(4):383–9.
160. Reus GZ, Fries GR, Stertz L, Badawy M, Passos IC, Barichello T, et al. The role of inflammation and microglial activation in the pathophysiology of psychiatric disorders. Neuroscience. 2015;300:141–54.
161. van Berckel BN, Bossong MG, Boellaard R, Kloet R, Schuitemaker A, Caspers E, et al. Microglia activation in recent-onset schizophrenia: a quantitative (R)-[11C]PK11195 positron emission tomography study. Biol Psychiatry. 2008;64(9):820–2.
162. Torres-Platas SG, Cruceanu C, Chen GG, Turecki G, Mechawar N. Evidence for increased microglial priming and macrophage recruitment in the dorsal anterior cingulate white matter of depressed suicides. Brain Behav Immun. 2014;42:50–9.
163. Frick LR, Williams K, Pittenger C. Microglial dysregulation in psychiatric disease. Clin Dev Immunol. 2013;2013:608654.
164. Paolicelli RC, Bolasco G, Pagani F, Maggi L, Scianni M, Panzanelli P, et al. Synaptic pruning by microglia is necessary for normal brain development. Science. 2011;333(6048):1456–8.
165. Parkhurst CN, Yang G, Ninan I, Savas JN, Yates JR 3rd, Lafaille JJ, et al. Microglia promote learning-dependent synapse formation through brain-derived neurotrophic factor. Cell. 2013;155(7):1596–609.
166. Tremblay ME, Lowery RL, Majewska AK. Microglial interactions with synapses are modulated by visual experience. PLoS Biol. 2010;8(11):e1000527.
167. Bloomfield PS, Selvaraj S, Veronese M, Rizzo G, Bertoldo A, Owen DR, et al. Microglial activity in people at ultra high risk of psychosis and in schizophrenia: an [(11)C]PBR28 PET brain imaging study. Am J Psychiatry. 2016;173(1):44–52.
168. Ligthart S, Vaez A, Vosa U, Stathopoulou MG, de Vries PS, Prins BP, et al. Genome analyses of >200,000 individuals identify 58 loci for chronic inflammation and highlight pathways that link inflammation and complex disorders. Am J Hum Genet. 2018;103(5):691–706.
169. Speer K, Upton D, Semple S, McKune A. Systemic low-grade inflammation in post-traumatic stress disorder: a systematic review. J Inflamm Res. 2018;11:111–21.
170. Jones KA, Thomsen C. The role of the innate immune system in psychiatric disorders. Mol Cell Neurosci. 2013;53:52–62.
171. Valkanova V, Ebmeier KP, Allan CL. CRP, IL-6 and depression: a systematic review and meta-analysis of longitudinal studies. J Affect Disord. 2013;150(3):736–44.
172. Courtet P, Jaussent I, Genty C, Dupuy AM, Guillaume S, Ducasse D, et al. Increased CRP levels may be a trait marker of suicidal attempt. Eur Neuropsychopharmacol. 2015;25(10):1824–31.
173. Cappi C, Muniz RK, Sampaio AS, Cordeiro Q, Brentani H, Palacios SA, et al. Association study between functional polymorphisms in the TNF-alpha gene and obsessive-compulsive disorder. Arq Neuropsiquiatr. 2012;70(2):87–90.
174. Denys D, Fluitman S, Kavelaars A, Heijnen C, Westenberg H. Decreased TNF-alpha and NK activity in obsessive-compulsive disorder. Psychoneuroendocrinology. 2004;29(7):945–52.
175. Konuk N, Tekin IO, Ozturk U, Atik L, Atasoy N, Bektas S, et al. Plasma levels of tumor necrosis factor-alpha and interleukin-6 in obsessive compulsive disorder. Mediat Inflamm. 2007;2007:65704.
176. Monteleone P, Catapano F, Fabrazzo M, Tortorella A, Maj M. Decreased blood levels of tumor necrosis factor-alpha in patients with obsessive-compulsive disorder. Neuropsychobiology. 1998;37(4):182–5.
177. Matthews KA, Schott LL, Bromberger JT, Cyranowski JM, Everson-Rose SA, Sowers M. Are there bi-directional associations between depressive symptoms and C-reactive protein in mid-life women? Brain Behav Immun. 2010;24(1):96–101.
178. Howren MB, Lamkin DM, Suls J. Associations of depression with C-reactive protein, IL-1, and IL-6: a meta-analysis. Psychosom Med. 2009;71(2):171–86.

179. Khandaker GM, Pearson RM, Zammit S, Lewis G, Jones PB. Association of serum interleukin 6 and C-reactive protein in childhood with depression and psychosis in young adult life: a population-based longitudinal study. JAMA Psychiat. 2014;71(10):1121–8.
180. Eraly SA, Nievergelt CM, Maihofer AX, Barkauskas DA, Biswas N, Agorastos A, et al. Assessment of plasma C-reactive protein as a biomarker of posttraumatic stress disorder risk. JAMA Psychiat. 2014;71(4):423–31.
181. Michopoulos V, Rothbaum AO, Jovanovic T, Almli LM, Bradley B, Rothbaum BO, et al. Association of CRP genetic variation and CRP level with elevated PTSD symptoms and physiological responses in a civilian population with high levels of trauma. Am J Psychiatry. 2015;172(4):353–62.
182. Miller AH, Raison CL. The role of inflammation in depression: from evolutionary imperative to modern treatment target. Nat Rev Immunol. 2016;16(1):22–34.
183. Maes M, Lambrechts J, Bosmans E, Jacobs J, Suy E, Vandervorst C, et al. Evidence for a systemic immune activation during depression: results of leukocyte enumeration by flow cytometry in conjunction with monoclonal antibody staining. Psychol Med. 1992;22(1):45–53.
184. Raison CL, Rutherford RE, Woolwine BJ, Shuo C, Schettler P, Drake DF, et al. A randomized controlled trial of the tumor necrosis factor antagonist infliximab for treatment-resistant depression: the role of baseline inflammatory biomarkers. JAMA Psychiat. 2013;70(1):31–41.
185. Dantzer R. Depression and inflammation: an intricate relationship. Biol Psychiatry. 2012;71(1):4–5.
186. Dowlati Y, Herrmann N, Swardfager W, Liu H, Sham L, Reim EK, et al. A meta-analysis of cytokines in major depression. Biol Psychiatry. 2010;67(5):446–57.
187. Krishnadas R, Cavanagh J. Depression: an inflammatory illness? J Neurol Neurosurg Psychiatry. 2012;83(5):495–502.
188. Dhabhar FS, Burke HM, Epel ES, Mellon SH, Rosser R, Reus VI, et al. Low serum IL-10 concentrations and loss of regulatory association between IL-6 and IL-10 in adults with major depression. J Psychiatr Res. 2009;43(11):962–9.
189. Muller N. Immunology of major depression. Neuroimmunomodulation. 2014;21(2–3):123–30.
190. Miller GE, Cole SW. Clustering of depression and inflammation in adolescents previously exposed to childhood adversity. Biol Psychiatry. 2012;72(1):34–40.
191. Leboyer M, Soreca I, Scott J, Frye M, Henry C, Tamouza R, et al. Can bipolar disorder be viewed as a multi-system inflammatory disease? J Affect Disord. 2012;141(1):1–10.
192. Kim HK, Andreazza AC, Elmi N, Chen W, Young LT. Nod-like receptor pyrin containing 3 (NLRP3) in the post-mortem frontal cortex from patients with bipolar disorder: a potential mediator between mitochondria and immune-activation. J Psychiatr Res. 2016;72:43–50.
193. Modabbernia A, Taslimi S, Brietzke E, Ashrafi M. Cytokine alterations in bipolar disorder: a meta-analysis of 30 studies. Biol Psychiatry. 2013;74(1):15–25.
194. Dargel AA, Godin O, Kapczinski F, Kupfer DJ, Leboyer M. C-reactive protein alterations in bipolar disorder: a meta-analysis. J Clin Psychiatry. 2015;76(2):142–50.
195. Mondelli V, Ciufolini S, Belvederi Murri M, Bonaccorso S, Di Forti M, Giordano A, et al. Cortisol and inflammatory biomarkers predict poor treatment response in first episode psychosis. Schizophr Bull. 2015;41(5):1162–70.
196. Miller BJ, Culpepper N, Rapaport MH. C-reactive protein levels in schizophrenia: a review and meta-analysis. Clin Schizophr Relat Psychoses. 2014;7(4):223–30.
197. Metcalf SA, Jones PB, Nordstrom T, Timonen M, Maki P, Miettunen J, et al. Serum C-reactive protein in adolescence and risk of schizophrenia in adulthood: a prospective birth cohort study. Brain Behav Immun. 2017;59:253–9.
198. Fernandes BS, Steiner J, Bernstein HG, Dodd S, Pasco JA, Dean OM, et al. C-reactive protein is increased in schizophrenia but is not altered by antipsychotics: meta-analysis and implications. Mol Psychiatry. 2016;21(4):554–64.

199. Passos IC, Vasconcelos-Moreno MP, Costa LG, Kunz M, Brietzke E, Quevedo J, et al. Inflammatory markers in post-traumatic stress disorder: a systematic review, meta-analysis, and meta-regression. Lancet Psychiatry. 2015;2(11):1002–12.
200. Cryan JF, Holmes A. The ascent of mouse: advances in modelling human depression and anxiety. Nat Rev Drug Discov. 2005;4(9):775–90.
201. Sanchez MM, Ladd CO, Plotsky PM. Early adverse experience as a developmental risk factor for later psychopathology: evidence from rodent and primate models. Dev Psychopathol. 2001;13(3):419–49.
202. Francis D, Diorio J, Liu D, Meaney MJ. Nongenomic transmission across generations of maternal behavior and stress responses in the rat. Science. 1999;286(5442):1155–8.
203. Harlow HF, Dodsworth RO, Harlow MK. Total social isolation in monkeys. Proc Natl Acad Sci U S A. 1965;54(1):90–7.
204. Hinde RA, Spencer-Booth Y. Effects of brief separation from mother on rhesus monkeys. Science. 1971;173(3992):111–8.
205. Levine S, Chevalier JA, Korchin SJ. The effects of early shock and handling on later avoidance learning. J Pers. 1956;24(4):475–93.
206. Suomi SJ. Early determinants of behaviour: evidence from primate studies. Br Med Bull. 1997;53(1):170–84.
207. Spitz RA. Hospitalism; an inquiry into the genesis of psychiatric conditions in early childhood. Psychoanal Study Child. 1945;1:53–74.
208. Bowlby J. Maternal care and mental health. Bull World Health Organ. 1951;3(3):355–533.
209. Kendler KS, Bulik CM, Silberg J, Hettema JM, Myers J, Prescott CA. Childhood sexual abuse and adult psychiatric and substance use disorders in women: an epidemiological and cotwin control analysis. Arch Gen Psychiatry. 2000;57(10):953–9.
210. Poole JC, Kim HS, Dobson KS, Hodgins DC. Adverse childhood experiences and disordered gambling: assessing the mediating role of emotion Dysregulation. J Gambl Stud. 2017;33(4): 1187–200.
211. Widom CS, DuMont K, Czaja SJ. A prospective investigation of major depressive disorder and comorbidity in abused and neglected children grown up. Arch Gen Psychiatry. 2007;64 (1):49–56.
212. Green JG, McLaughlin KA, Berglund PA, Gruber MJ, Sampson NA, Zaslavsky AM, et al. Childhood adversities and adult psychiatric disorders in the national comorbidity survey replication I: associations with first onset of DSM-IV disorders. Arch Gen Psychiatry. 2010;67(2):113–23.
213. Kessler RC, Davis CG, Kendler KS. Childhood adversity and adult psychiatric disorder in the US National Comorbidity Survey. Psychol Med. 1997;27(5):1101–19.
214. McLaughlin KA, Greif Green J, Gruber MJ, Sampson NA, Zaslavsky AM, Kessler RC. Childhood adversities and first onset of psychiatric disorders in a national sample of US adolescents. Arch Gen Psychiatry. 2012;69(11):1151–60.
215. Schaefer JD, Moffitt TE, Arseneault L, Danese A, Fisher HL, Houts R, et al. Adolescent victimization and early-adult psychopathology: approaching causal inference using a longitudinal twin study to rule out noncausal explanations. Clin Psychol Sci. 2018;6(3):352–71.
216. Gilbert R, Kemp A, Thoburn J, Sidebotham P, Radford L, Glaser D, et al. Recognising and responding to child maltreatment. Lancet. 2009;373(9658):167–80.
217. Varese F, Smeets F, Drukker M, Lieverse R, Lataster T, Viechtbauer W, et al. Childhood adversities increase the risk of psychosis: a meta-analysis of patient-control, prospective-and cross-sectional cohort studies. Schizophr Bull. 2012;38(4):661–71.
218. Widom CS. Posttraumatic stress disorder in abused and neglected children grown up. Am J Psychiatry. 1999;156(8):1223–9.
219. Mueller SC, Hardin MG, Korelitz K, Daniele T, Bemis J, Dozier M, et al. Incentive effect on inhibitory control in adolescents with early-life stress: an antisaccade study. Child Abuse Negl. 2012;36(3):217–25.

220. Goff B, Gee DG, Telzer EH, Humphreys KL, Gabard-Durnam L, Flannery J, et al. Reduced nucleus accumbens reactivity and adolescent depression following early-life stress. Neuroscience. 2013;249:129–38.
221. Mehta MA, Gore-Langton E, Golembo N, Colvert E, Williams SC, Sonuga-Barke E. Hyporesponsive reward anticipation in the basal ganglia following severe institutional deprivation early in life. J Cogn Neurosci. 2010;22(10):2316–25.
222. Brown GWHT. Social origins of depression: a study of psychiatric disorder in women. New York: The Free Press; 1978.
223. Danese A, Moffitt TE, Harrington H, Milne BJ, Polanczyk G, Pariante CM, et al. Adverse childhood experiences and adult risk factors for age-related disease: depression, inflammation, and clustering of metabolic risk markers. Arch Pediatr Adolesc Med. 2009;163(12):1135–43.
224. Agnew-Blais J, Danese A. Childhood maltreatment and unfavourable clinical outcomes in bipolar disorder: a systematic review and meta-analysis. Lancet Psychiatry. 2016;3(4):342–9.
225. Maercker A, Brewin CR, Bryant RA, Cloitre M, van Ommeren M, Jones LM, et al. Diagnosis and classification of disorders specifically associated with stress: proposals for ICD-11. World Psychiatry. 2013;12(3):198–206.
226. Galea I, Bechmann I, Perry VH. What is immune privilege (not)? Trends Immunol. 2007;28 (1):12–8.
227. Yirmiya R, Goshen I. Immune modulation of learning, memory, neural plasticity and neurogenesis. Brain Behav Immun. 2011;25(2):181–213.
228. Gillespie CFGS, Binder EB, Schatzberg AF, Nemeroff CB. In: Schatzberg AF, Nemeroff CB, editors. Textbook of psychopharmacology. New York: America Psychiatric Publishing; 2009.
229. Maes M, Leonard BE, Myint AM, Kubera M, Verkerk R. The new '5-HT' hypothesis of depression: cell-mediated immune activation induces indoleamine 2,3-dioxygenase, which leads to lower plasma tryptophan and an increased synthesis of detrimental tryptophan catabolites (TRYCATs), both of which contribute to the onset of depression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2011;35(3):702–21.
230. Raison CL, Dantzer R, Kelley KW, Lawson MA, Woolwine BJ, Vogt G, et al. CSF concentrations of brain tryptophan and kynurenines during immune stimulation with IFN-alpha: relationship to CNS immune responses and depression. Mol Psychiatry. 2010;15 (4):393–403.
231. Tavares RG, Tasca CI, Santos CE, Alves LB, Porciuncula LO, Emanuelli T, et al. Quinolinic acid stimulates synaptosomal glutamate release and inhibits glutamate uptake into astrocytes. Neurochem Int. 2002;40(7):621–7.
232. Hardingham GE, Fukunaga Y, Bading H. Extrasynaptic NMDARs oppose synaptic NMDARs by triggering CREB shut-off and cell death pathways. Nat Neurosci. 2002;5(5):405–14.
233. Chao CC, Hu S, Molitor TW, Shaskan EG, Peterson PK. Activated microglia mediate neuronal cell injury via a nitric oxide mechanism. J Immunol. 1992;149(8):2736–41.
234. Kehrer JP. Cause-effect of oxidative stress and apoptosis. Teratology. 2000;62(4):235–6.
235. Uttara B, Singh AV, Zamboni P, Mahajan RT. Oxidative stress and neurodegenerative diseases: a review of upstream and downstream antioxidant therapeutic options. Curr Neuropharmacol. 2009;7(1):65–74.
236. Beal MF. Bioenergetic approaches for neuroprotection in parkinson's disease. Ann Neurol. 2003;53(Suppl 3):S39–47. discussion S-8
237. Christen Y. Oxidative stress and Alzheimer disease. Am J Clin Nutr. 2000;71(2):621S.
238. Mahadik SP, Mukherjee S. Free radical pathology and antioxidant defense in schizophrenia: a review. Schizophr Res. 1996;19(1):1–17.
239. Perry VH, Newman TA, Cunningham C. The impact of systemic infection on the progression of neurodegenerative disease. Nat Rev Neurosci. 2003;4(2):103–12.
240. Breder CD, Hazuka C, Ghayur T, Klug C, Huginin M, Yasuda K, et al. Regional induction of tumor necrosis factor alpha expression in the mouse brain after systemic lipopolysaccharide administration. Proc Natl Acad Sci U S A. 1994;91(24):11393–7.

241. Monje ML, Toda H, Palmer TD. Inflammatory blockade restores adult hippocampal neurogenesis. Science. 2003;302(5651):1760.
242. Weaver IC, Cervoni N, Champagne FA, D'Alessio AC, Sharma S, Seckl JR, et al. Epigenetic programming by maternal behavior. Nat Neurosci. 2004;7(8):847–54.
243. Klengel T, Mehta D, Anacker C, Rex-Haffner M, Pruessner JC, Pariante CM, et al. Allele-specific FKBP5 DNA demethylation mediates gene-childhood trauma interactions. Nat Neurosci. 2013;16(1):33–41.
244. Miller GE, Chen E. Harsh family climate in early life presages the emergence of a proinflammatory phenotype in adolescence. Psychol Sci. 2010;21(6):848–56.
245. Gregor MF, Hotamisligil GS. Inflammatory mechanisms in obesity. Annu Rev Immunol. 2011;29:415.
246. Gregory AM, Sadeh A. Annual research review: sleep problems in childhood psychiatric disorders—a review of the latest science. J Child Psychol Psychiatry. 2016;57(3):296–317.
247. Kajeepeta S, Gelaye B, Jackson CL, Williams MA. Adverse childhood experiences are associated with adult sleep disorders: a systematic review. Sleep Med. 2015;16(3):320–30.
248. Noll JG, Trickett PK, Susman EJ, Putnam FW. Sleep disturbances and childhood sexual abuse. J Pediatr Psychol. 2006;31(5):469–80.
249. Irwin MR, Wang M, Campomayor CO, Collado-Hidalgo A, Cole S. Sleep deprivation and activation of morning levels of cellular and genomic markers of inflammation. Arch Intern Med. 2006;166(16):1756–62.
250. Miller MA, Kandala NB, Kivimaki M, Kumari M, Brunner EJ, Lowe GD, et al. Gender differences in the cross-sectional relationships between sleep duration and markers of inflammation: Whitehall II study. Sleep. 2009;32(7):857–64.
251. Felger JC, Mun J, Kimmel HL, Nye JA, Drake DF, Hernandez CR, et al. Chronic interferon-alpha decreases dopamine 2 receptor binding and striatal dopamine release in association with anhedonia-like behavior in nonhuman primates. Neuropsychopharmacology. 2013;38(11):2179–87.
252. Dowell NG, Cooper EA, Tibble J, Voon V, Critchley HD, Cercignani M, et al. Acute changes in striatal microstructure predict the development of interferon-alpha induced fatigue. Biol Psychiatry. 2016;79(4):320–8.
253. Haroon E, Woolwine BJ, Chen X, Pace TW, Parekh S, Spivey JR, et al. IFN-alpha-induced cortical and subcortical glutamate changes assessed by magnetic resonance spectroscopy. Neuropsychopharmacology. 2014;39(7):1777–85.
254. Harrison NA, Cercignani M, Voon V, Critchley HD. Effects of inflammation on hippocampus and substantia nigra responses to novelty in healthy human participants. Neuropsychopharmacology. 2015;40(4):831–8.
255. Harrison NA, Voon V, Cercignani M, Cooper EA, Pessiglione M, Critchley HD. A Neurocomputational account of how inflammation enhances sensitivity to punishments versus rewards. Biol Psychiatry. 2016;80(1):73–81.
256. Harrison NA, Brydon L, Walker C, Gray MA, Steptoe A, Dolan RJ, et al. Neural origins of human sickness in interoceptive responses to inflammation. Biol Psychiatry. 2009;66(5):415–22.
257. Slavich GM, Way BM, Eisenberger NI, Taylor SE. Neural sensitivity to social rejection is associated with inflammatory responses to social stress. Proc Natl Acad Sci U S A. 2010;107(33):14817–22.
258. Eisenberger NI, Lieberman MD. Why rejection hurts: a common neural alarm system for physical and social pain. Trends Cogn Sci. 2004;8(7):294–300.
259. Muscatell KA, Dedovic K, Slavich GM, Jarcho MR, Breen EC, Bower JE, et al. Greater amygdala activity and dorsomedial prefrontal-amygdala coupling are associated with enhanced inflammatory responses to stress. Brain Behav Immun. 2015;43:46–53.
260. Inagaki TK, Muscatell KA, Irwin MR, Cole SW, Eisenberger NI. Inflammation selectively enhances amygdala activity to socially threatening images. NeuroImage. 2012;59(4):3222–6.

261. Bilbo SD, Schwarz JM. Early-life programming of later-life brain and behavior: a critical role for the immune system. Front Behav Neurosci. 2009;3:14.
262. Knuesel I, Chicha L, Britschgi M, Schobel SA, Bodmer M, Hellings JA, et al. Maternal immune activation and abnormal brain development across CNS disorders. Nat Rev Neurol. 2014;10(11):643–60.
263. Patterson PH. Immune involvement in schizophrenia and autism: etiology, pathology and animal models. Behav Brain Res. 2009;204(2):313–21.
264. Short SJ, Lubach GR, Karasin AI, Olsen CW, Styner M, Knickmeyer RC, et al. Maternal influenza infection during pregnancy impacts postnatal brain development in the rhesus monkey. Biol Psychiatry. 2010;67(10):965–73.
265. Khandaker GM, Zimbron J, Lewis G, Jones PB. Prenatal maternal infection, neurodevelopment and adult schizophrenia: a systematic review of population-based studies. Psychol Med. 2013;43(2):239–57.
266. Mednick SA, Machon RA, Huttunen MO, Bonett D. Adult schizophrenia following prenatal exposure to an influenza epidemic. Arch Gen Psychiatry. 1988;45(2):189–92.
267. Danese A, McCrory E. Child maltreatment. In: Rutter's textbook of child & adolescent psychiatry. 6th ed. London: Wiley-Blackwell; 2015.

# C-Reactive Protein (CRP): A Potent Inflammation Biomarker in Psychiatric Disorders

7

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**Abstract**

An increasing number of studies have investigated the role of inflammation in psychiatric disorders, by demonstrating how an altered/dysfunctional immunological and inflammatory system may underpin a psychiatric condition. Particularly, several studies specifically investigated the role of a neuroinflammatory biomarker, named C-reactive protein (CRP), in psychiatric disorders. Overall, even though scientific literature so far published still does not appear definitive, CRP is more likely reported to be elevated in several psychiatric disorders, including schizophrenia, mood disorders, anxiety disorders and post-traumatic stress disorder. Moreover, a low-grade inflammation (CRP >3 mg/L) has been more likely observed in a subgroup of patients affected with a more severe psychopathological symptomatology, more treatment resistance and worst clinical mental illness course, strengthening the hypothesis of the need for a different clinical and prognostic characterization based on this concomitant neuroinflammatory predisposition. However, even though further research studies are needed to confirm this preliminary evidence, CRP may represent a potential clinical routine biomarker which could be integrated in the clinical routine practice to better characterize clinical picture and course as well as address clinicians towards a personalized treatment.

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**Keywords**

Biomarkers · C-Reactive protein · CRP; Immuno-modulation · Immuno-psychiatry · Inflammatory markers · Mental health · Neuroinflammation · Psychiatry disorders

## 7.1 Introduction

C-reactive protein (CRP) is a pentameric acute-phase reactant protein, member of the pentraxin family, produced mainly by the hepatocytes, as a result of the activation of the innate humoral system. The increasing CRP level is stimulated by the production of a set of inflammatory cytokines, especially interleukin-6 (IL-6), enhanced synergically by IL-1β, which are secreted by macrophages and T cells during the acute phase of an inflammatory process [1–3]. CRP was firstly identified in the serum samples from patients affected by *Streptococcus pneumoniae* in 1930 [4]. The name CRP was derived by its reaction with the capsular polysaccharide antigen of the bacteria [4]. The human CRP gene is located at 1q23.2 on the long arm of chromosome 1 and it is produced as a native protein (nCRP), which can irreversibly dissociate itself into five separate monomers (mCRP) at the inflammation and infection sites. These two types of isomers display different antigenic, biological and electrophoretic activities [2, 5]. The role of CRP is to recognize and eliminate pathogens as well as damage cells, by binding itself at various ligands, such as lysophosphatidylcholine, phospholipids, histone, chromatin and fibronectin, by activating the complement system, by binding at Fc receptors and by activating several other inflammatory-related mechanisms [3]. CRP has both pro-inflammatory and anti-inflammatory properties. In fact, nCRP may exert anti-inflammatory activities, mainly by inducing phagocytosis, promoting apoptosis and limiting the generation of the membrane attack complex (MAC) and C5a, or by suppressing the adherence of platelets to neutrophils. Conversely, mCRP may promote monocyte chemotaxis, the recruitment of circulating leukocytes towards the inflammatory sites, by increasing IL-8 and MCP-1 cytokine production [2, 6]. However, the precise mechanism by which CRP interacts within the immune response is not entirely understood.

In clinical practice, CRP is generally used as a biomarker of infection, chronic disease state and chronic low-grade inflammation [7]. A normal value of CRP levels is considered below 3 mg/dL, although levels between 1 mg/dL and 10 mg/dL are considered borderline levels which should be accurately investigated [3]. Generally, CRP levels rise and fall rapidly with the onset and removal of the inflammatory stimulus [8]. Moreover, CRP is correlated with cardiovascular risk, including myocardial infarction, stroke, sudden cardiovascular death and peripheral vascular disease and its levels may change depending on the occurrence of the abovementioned clinical conditions [9]. The high-sensitivity CRP (hs-CRP) is easily measured through a blood sample [8]. In general, hs-CRP levels ranging 1–3 mg/L and greater than 3 mg/L are respectively associated with a moderate and high risk for

the development of a cardiovascular disease [8, 10]. Furthermore, baseline hs-CRP levels may hugely vary across different individuals, depending on their inflammatory state, and these levels may also vary whether an individual is taking some types of medications (e.g. non-steroidal anti-inflammatory drugs [NSAIDS], statins, etc.) [3].

Overall, recently, a wide range of scientific evidence is bringing to light on how inflammation and, more generally, the immunity dysregulation may play a crucial role in psychiatric disorders [11–20]. In particular, several mechanisms have been hypothesized by which CRP may interact with the central nervous system (CNS), either via indirect effects through peripheral signalling or through more direct central effects [17]. Among these supposed mechanisms, an increased dysregulated activation of the complement pathway (mediated by CRP) has been observed in some psychiatric disorders, such as schizophrenia and depression [15, 21]. Furthermore, increased CRP levels and its pro-inflammatory activity which may drive a CNS inflammation, through microglia and astrocytes activation, have been observed in several psychiatric disorders [22–24]. Moreover, it has been hypothesized that peripheral myeloid cells or pro-inflammatory cytokines can induce a neurovascular damage and an upregulation of the matrix metalloproteinases (MMPs), by increasing the blood–brain barrier (BBB) permeability [25, 26]. CRP usually does not freely cross the BBB [27]. An increased BBB permeability may be determined by a severe stress and/or a traumatic brain injury which indeed may facilitate an easy access of peripheral CRP through the BBB into the cerebrospinal fluid (CSF) [26, 28]. Similarly, CRP may in turn induce BBB disruption, through the binding and activation of Fc gamma receptors (expressed also in the microglia and astrocytes), CD16 and CD32 present on the endothelial cells [29].

With this in mind, in the paragraphs below, a more in-depth overview will be carried out on how CRP may play a clinically relevant role in different psychiatric disorders and whether CRP levels may be associated with more severe and treatment-resistant psychopathological states.

## 7.2 Depressive Disorders

Depression is the most common mental illness, affecting around 10–20% of the general population, by representing one of the leading causes of disability worldwide [30]. The aetiopathogenesis of depression is highly complex and not entirely understood. The most widely accepted pathophysiology of depression is based on the monoaminergic theory [31], although recent research directions focused on other pathways, such as genetic susceptibility and epigenetic modifications, the dysregulation of the hypothalamus–pituitary–adrenal axis (HPA), hippocampal and frontal lobe dysfunction, oxidative stress–induced damage and the neurodevelopment theory of depression which pointed out on the risk/protective factors occurring at the earlier stages of human life, including the prenatal period [32–36].

Furthermore, recent studies have also hypothesized the role of immune dysregulation in the aetiopathogenesis of depression [37, 38], by supporting the evidence about the association between inflammation and depression [39–42]. In this regard, a pro-inflammatory state encountered during inflammatory diseases has been associated with the so-called sickness behaviour characterized by a depression-like symptomatology, such as anhedonia, weight and appetite loss, memory impairment and cognitive and social dysfunction, which can be frequently reported in major depressive disorder (MDD) [43]. In this regard, several studies investigated the role of CRP levels in depressive individuals, by mainly observing increased CRP levels [38, 42, 44], even though findings are often contrasting [16, 45]. A retrospective cohort study reported that elevated serum hs-CRP levels in women may indeed represent an independent risk factor for de novo major depressive disorder [46]. CRP levels have been reported to be higher in atypical major depressive disorder (MDD) compared to other MDD variants [47], in MDD patients with predominantly somatic symptomatology [48] and in MDD patients at higher risks of psychiatric hospitalization [44]. Moreover, higher levels of CRP and IL-6 at baseline predicted the risk of persistent depressive symptoms over 5 years [49] and concomitant cognitive symptomatology during a 12-year follow-up [50]. In addition, higher hs-CRP levels were significantly associated with a more severe MDD symptomatology, particularly among women who also reported concomitant cognitive symptomatology and suicidality [51]. Higher CRP levels have been also observed in treatment-resistant MDD patients (TRD) [52] and in those MDD individuals who are early responders to paroxetine treatment [53]. Conversely, other studies reported lower baseline CRP levels among MDD patients who display a better and faster response to SSRI treatment [51, 54].

Overall, about a third of all depressed patients seem to display elevated serum CRP (>3 mg/L) indicating a low-grade inflammatory state [44, 55, 56]. A chronic low-grade inflammation may be associated with a different MDD subgroup with a distinct aetiopathogenesis, different clinical course, treatment response and prognosis [37, 51]. In fact, one could argue that those MDD patients who do not adequately respond to treatment should be investigated regarding a concomitant pro-inflammatory state [52, 57, 58]. Furthermore, considering the anti-inflammatory properties of selective serotonin reuptake inhibitors (SSRIs), selective serotonin and noradrenaline inhibitors (SNRIs) and tricyclic antidepressant (TCAs), one could hypothesize a possible additional mechanism of antidepressants through which they can indirectly reduce depressive symptoms by acting on the inflammatory state [59–61]. Finally, based on the inflammatory hypothesis, anti-inflammatory drugs (e.g. non-steroidal anti-inflammatory drugs [NSAIDs] or anti-cytokine) could be helpful in treating depression [62–66]. However, findings are so far contradictory, being reported a beneficial effect only in those patients with CRP levels higher than 5 mg/L [67] or not observing any clinically relevant improvement in depressive symptomatology [68].

Contrasting findings have been published regarding the association between CRP and postpartum depression (PPD) [69–72]. For example, Roomruangwong et al. [73] observed that increased CRP levels in the third trimester were strongly associated

with depressive symptomatology in the prenatal and early postpartum period. Similarly, higher hs-CRP and IL-6 levels have been found significantly associated with the onset of PPD at 6 months [69, 74]. Long-term studies evaluating newborns of mothers with elevated CRP levels during pregnancy found that higher maternal CRP levels, particularly during the third trimester, may predict poorer child cognitive flexibility in the third trimester [75], the development of ADHD particularly in boys [76, 77], schizophrenia [78] and autism [79]. However, these findings are still controversial and not definitive and should be further investigated to clarify the role of CRP in pregnancy and the postpartum period [80, 81].

Therefore, further larger and methodological homogeneous studies should be carried out in order to better understand the role of CRP levels in MDD and PPD.

## 7.3 Bipolar Disorders

Bipolar disorder (BD) is a mood disorder characterized by recurring mood states, ranging from mania/hypomania to mild–moderate–severe depression, interspersed with period(s) of euthymia [82]. BD is associated with functional impairment, high disability, healthcare costs, premature mortality and increased risk of cardiovascular disease compared to the general population [13, 83–85].

Despite the pathogenesis of BD being widely studied [86, 87], some research directions suggested a possible role of the immune system in BD aetiopathogenesis [88, 89]. In fact, BD patients are usually accompanied by high physical comorbidity involving the immune system (e.g. cardiovascular diseases and autoimmune disorders such as diabetes mellitus, autoimmune thyroiditis, systemic lupus erythematosus, psoriasis and inflammatory bowel disease) [13]. Furthermore, altered levels of inflammatory mediators, such as the cytokine system, have been reported in BD individuals [41, 85, 90], including altered CRP levels [91–93]. CRP levels have been significantly reported higher in manic and euthymic BD phases but not in depressed phases [91], by suggesting that CRP may represent an early warning sign for the onset of a manic phase in depressed BD individuals [92, 94–96]. A prospective study reported significant higher CRP levels in depressed BD II compared to unipolar MDD individuals, suggesting that CRP may represent a useful biomarker to differentiate between MDD and BD II depression in both their depressed and euthymic state [97]. A population-based study revealed increased CRP levels in individuals at higher risk of late-onset BD [98]. Increased CRP levels have been also observed in more severe BD patients [99–101]. A recent systematic review and meta-analysis [85] showed higher pro-inflammatory immune biomarkers, including increased CRP levels, in BD patients compared to the control group. The meta-regression analysis reported a marginally significant inverse and negligible association between CRP levels and BD phase duration, by suggesting a possible role of CRP as a biomarker of mood episodes within BD [85].

Although few studies investigated the role of CRP genetic polymorphism in BD individuals [102, 103], a significantly higher prevalence of the CRP rs1130864 A

allele has been observed in those BD patients with a concomitant thyroid disorder and in those BD patients with a rapid cycling illness course [102].

Furthermore, few studies investigated the association between CRP levels and cognitive impairment in BD individuals [104, 105], by observing a worst cognitive performance in those BD patients who displayed CRP levels greater than 5 mg/L compared to those with lower CRP levels [106]. However, further studies are needed to confirm this preliminary evidence.

Although mood stabilizers may exert an anti-inflammatory activity [61, 107], there is still poor literature about the association between CRP levels and mood stabilizer treatment [91, 103, 108–110]. Similarly, few studies investigating the role of anti-inflammatory and/or neuromodulator drugs in the improvement of BD reported inconclusive results [111]. Therefore, although some findings are promising, results are still controversial and further studies should be carried out in order to evaluate whether CRP may represent a sensitive and specific biomarker of BD phases and/or recrudescence, severity, etc.

## 7.4 Suicidality

Overall, the role of CRP in suicidal ideation and/or behaviour has been investigated, reporting a significant association with history of suicidal attempts in depressed patients [112, 113], a greater risk of suicide after 9 years of follow-up [114] and suicidal ideation [115]. Moreover, elevated CRP levels have been associated with anger, hostility, impulsivity and aggressiveness [116–118] which may be indirectly related to a higher probability of suicidality.

Few studies evaluated the role of inflammation-related genes in suicidal behaviour, by reporting a significant association between a polymorphism located at the CRP gene, +1444C > T (rs1130864), and a predisposition to trait impulsiveness in women [119] and between +1444 T allele and suicide attempters compared to +1444C allele [120].

## 7.5 Schizophrenia and Psychotic Spectrum Disorders

Schizophrenia is a complex psychiatric disorder characterized by positive and negative symptoms, cognitive deficits and a gradual functional impairment, with a prevalence of around 1% in the general population [9, 121]. The aetiopathogenesis of schizophrenia and psychotic spectrum disorders is multi-determined by the reciprocal interactions between genetic, environmental and social determinants [122–124]. Furthermore, a potential determinant role of the immunological and inflammatory system has also been supposed, by observing a low-grade inflammatory state in schizophrenia and psychotic spectrum disorders [125–127]. In schizophrenia individuals, several altered immunological and pro-inflammatory cytokines, as well as a higher prevalence of positive antinuclear antibodies and modifications in white blood cell (WBC) count, have been reported, by supporting the hypothesis of a

potential role of the immunological system in the pathogenesis of schizophrenia [127–129]. Moreover, schizophrenia individuals usually display parameters more likely associated to a pro-inflammatory state, such as high BMI, metabolic syndrome, smoking status and autoimmune disorders (e.g. psoriasis, celiac disease and pernicious anaemia) [130–133]. Furthermore, as altered inflammatory markers have been found since the prodromal stage and in first psychotic episode (FEP) of schizophrenia, a potential role of the inflammation in the aetiopathogenesis of schizophrenia has been hypothesized, at least in a subgroup of schizophrenia patients [134, 135].

Within this context, some studies investigated the role of CRP as a biomarker of schizophrenia and psychotic spectrum disorders, by reporting baseline CRP levels greater than 3 mg/L in those adolescents who more likely will develop schizophrenia [55, 136]; in those schizophrenia patients with an acute exacerbation, compared to the healthy control group and chronic schizophrenia individuals [121, 137]; in those schizophrenia individuals who manifest a late or very-late-onset schizophrenia [44]; and in those patients who manifest more negative and severe psychotic symptomatology [138, 139], even though another study reported higher CRP levels in those who manifest more severe positive versus negative symptomatology [92].

Recent genetic studies confirmed the role of inflammation in the development of schizophrenia and psychotic spectrum disorders, by identifying two genetic risk scores, i.e. the presence of four single nucleotide polymorphisms (SNPs) in the CRP gene and the presence of 18 SNPs which appear to be more likely associated with higher CRP levels in a largest genome-wide association study (GWAS) carried out in schizophrenic individuals [66, 140].

Furthermore, only few studies investigate the association between CRP levels and cognitive impairment in schizophrenia individuals, by mainly reporting a significant association with an impaired semantic and working memory, general intellectual ability, abstract reasoning, an impaired mental flexibility and processing speed and poor verbal abilities and attention [104, 121, 141–145]. Moreover, higher CRP levels as well as an inverse association have been observed between cortical thickness in the frontal, insula and temporal brain regions and CRP levels [121].

Although antipsychotics may partially normalize immune alterations in schizophrenia individuals [145–147], a meta-analysis showed how increased CRP levels in schizophrenic patients do not necessarily change following an antipsychotic treatment [92]. Recently, few studies reported promising findings following clozapine treatment in significantly reducing WBC, IL-6 and CRP levels [127, 148] and aripiprazole [149]. Similarly, few studies investigated the potential role of NSAID and/or anti-inflammatory drug augmentation strategy in schizophrenia [150, 151], even though a recent meta-analysis reported a significant efficacy only with aspirin, oestrogens, minocycline and N-acetylcysteine, particularly in FEP and early-onset schizophrenia [152]. Therefore, although some findings are promising, results are still controversial and further studies should be carried out in order to evaluate whether CRP may represent a useful biomarker in schizophrenia and psychotic spectrum disorders.

## 7.6 Fear and Anxiety Disorders

Fear and anxiety disorders include general anxiety disorder (GAD), phobic disorders (PD), agoraphobia, social phobia and specific phobias, according to the DSM-5 [153]. It has been largely demonstrated how the inflammatory system may play a significant role in the pathogenesis of several fear and anxiety disorders, such as how the exposure to traumatic and stressful life events may determine the activation of the HPA axis, the immune system and the subsequent release of a set of pro-inflammatory cytokines [12]. However, very few studies reported significant findings about CRP, which was observed more elevated in a sample of individuals with a non-specific anxious state [154], in GAD children and adolescents [155] and in a male sample affected by mixed anxiety disorder [156], compared to a healthy control group. Conversely, no significant CRP differences have been reported in individuals with agoraphobia [157] and in another case-control study recruiting anxious individuals [158], compared to the healthy control group. A recent systematic review [159] reported significantly higher CRP levels in five studies recruiting GAD patients [156, 160–163], with only one study reporting an inverse correlation between CRP levels and GAD [164]. Finally, a population-based study reported a significant association between increased CRP levels and a diagnosis of panic disorder with agoraphobia [165]. Overall, there are still limiting and contrasting findings regarding the potential role of CRP in fear and anxiety disorders to be able to draw up conclusive evidence. Further research is needed to better understand new research directions in this regard.

## 7.7 Post-Traumatic Stress Disorder (PTSD)

Post-traumatic stress disorder (PTSD) is a mental disease which may generally occur after an objective traumatic event or a subjunctive trigger stimulus, characterized by highly intrusive memories, flashbacks, nightmares, avoidant behaviours related to traumatic cues, increased arousal or hypervigilance [166]. PTSD may be accompanied with comorbid MDD, substance use disorder (SUD) and suicidal ideation and/or attempt [12].

Overall, it has been well documented how being exposed to a trauma may be associated with a pro-inflammatory activity, the dysregulation of the HPA axis, alteration in the immune cell system and increased IL-6, TNF-α, INF-γ and CRP levels [12, 167–170]. Moreover, individuals who have been exposed to childhood maltreatment or difficult familial, socio-economic circumstances during their childhood reported significantly higher CRP levels in their adulthood, compared to those not exposed in childhood [171–173]. Elevated CRP levels have been reported in PTSD patients [174–180], particularly in those PTSD patients who manifest severe symptomatology, concomitant dissociative and/or depressive symptoms as well as more avoidance and re-experiencing domains [12, 78, 173, 179, 181]. However, other studies reported conflicting results [182].

Furthermore, altered immune gene expression and methylation patterns have also been associated with PTSD [166, 183]. In particular, rs1130864, a SNP in CRP gene expression, has been associated with increased peripheral CRP levels in more severe PTSD individuals and with a higher probability to develop PTSD in those traumatized individuals [184]. Moreover, PTSD was associated with higher CRP levels mediated by SNPs and methylation of the CRP gene promoter locus AIM2 in a sample of military veterans of post-9/11 events [185]. Therefore, further studies should be carried out in order to evaluate whether CRP may represent a useful biomarker in PTSD, whether it may correlate with severity and which PTSD-related domains and which treatment strategies may be further investigated and implemented considering this confirmed association between inflammatory state, CRP levels and PTSD.

## 7.8 Obsessive-Compulsive Disorder (OCD)

Although the aetiopathogenesis of obsessive-compulsive disorder (OCD) is still not completely understood and several multifactorial hypotheses have been proposed, there are still few studies investigating the role of inflammation in OCD, mainly focussing on the potential role of oxidative stress and free radicals in the brain tissues as well as in the immunological dysfunction occurring in a subset of OCD individuals [186, 187]. In particular, a persistent low-grade inflammation in OCD individuals has been documented, which could be determined by a pre-existing immuno-genetic susceptibility which may also explain the higher prevalence of autoimmune diseases, such as Sjögren's syndrome and celiac disease, Guillain–Barré syndrome, Crohn's disease, Hashimoto's thyroiditis and type 1 diabetes mellitus, in OCD patients [188, 189]. However, a recent meta-analysis did not report any significant differences in the levels of TNF-α, IL-6, IL-1β, IL-4, IL-10 or IN-γ between individuals with OCD and healthy control [190].

Overall, few studies investigated the role of CRP in OCD individuals, by demonstrating that elevated CRP levels have been associated with a poor insight, earliest age of OCD onset, higher suicidality and a family history of OCD [19, 20, 187, 191]. Overall, there are still limiting findings regarding the potential role of CRP in OCD which should address further research directions to be implemented in this regard.

## 7.9 Attention Deficit Hyperactivity Disorder (ADHD) and Autism Spectrum Disorder (ASD)

Attention deficit hyperactivity disorder (ADHD) represents the most common neurodevelopmental disorder in children, characterized by four core domains (i.e. inattention, hyperactivity, impulsivity and destruction) [192, 193]. Although it has been demonstrated to have a strong genetic component, recent studies revealed how immunological mechanisms and oxidative stress may be involved in the

aetiopathogenesis of ADHD [193–200]. In fact, ADHD is frequently associated with comorbid immunological conditions, such as atopic dermatitis, type 1 diabetes, hypothyroidism and asthma, which may suggest an immune-mediated pathway [200, 201].

Overall, few studies specifically investigated the role of CRP in ADHD, by reporting elevated CRP and IL-6 levels associated with low bedtime salivary cortisol, TNF-α and brain-derived neurotrophic factors (BDNF) in ADHD youths [143], elevated CRP levels associated with poor emotional regulation and more severe impulse dyscontrol [193, 201]. However, other studies reported contrasting findings, mainly due to heterogeneous methodologies and small sample size recruited [197, 202].

Autism spectrum disorder (ASD) is often a comorbid condition with ADHD [201, 203]. Few studies investigating the role of CRP in ASD, being only reported in a recent meta-analysis [204], reported significantly elevated CRP levels in peripheral blood in ASD children compared to healthy controls.

Overall, there are still limiting and contrasting findings regarding the potential role of CRP in neurodevelopmental disorders. Therefore, further studies are needed.

## 7.10 Addictive Disorders

Several studies are recently investigating the relationship between substance and/or alcohol use disorders (SUD and AUD) and inflammation [205–207]. For example, alcohol consumption may provoke an elevation of pro-inflammatory cytokines, such as IL-6, IL-10, IL-12, INF-γ and TNF-α [208–211] and the immune system may be involved in the development of alcohol hangover [212–214]. An analysis of the National Health and Nutrition Examination Survey (NHNES) reported lower CRP levels in past or current marijuana users, compared to non-marijuana users, by supporting the hypothesis of a potential anti-inflammatory activity of cannabis, even though prospective studies demonstrated that elevated CRP levels may indeed predict cannabis use and nicotine dependence [207, 215–219]. Elevated CRP levels have been reported in cocaine, tobacco, opioid and alcohol users [192, 205, 207, 216, 220–222]. However, further studies are needed to better understand the association between opioid use and CRP levels as other studies reported an anti-inflammatory effect of opioids [223–225]. For example, a study investigating the CRP levels in OUD patients undergoing methadone maintenance treatment (MMT) found significantly lower CRP levels after a 12-week MMT [206]. Inconclusive findings have been found between CRP and cocaine use disorder [226]. Therefore, further studies should be carried out in order to better understand whether CRP may be helpful as biomarkers in SUD and/or AUD.

## 7.11 COVID-19-Related Psychopathology

As the current COVID-19-related pandemic significantly increased the emergence of studies specifically addressed on inflammatory and immune system impairment in patients affected by COVID-19, it would be interesting to evaluate if any study deepened COVID-19-related inflammatory status and psychiatric disorders, as patients affected by COVID-19 frequently manifested de novo depressive disorders, PTSD, OCD and anxiety disorders or recurrence of previously diagnosed psychiatric conditions [227–229]. COVID-19 infection may induce an overproduction of pro-inflammatory cytokines which could be indirectly related to neuropsychiatric symptomatology onset [228, 229]. Furthermore, within the context of COVID-19 infection, an increase in CRP concentration is often described [228, 230] and it may be related with the development of severe COVID-19 disease and poor prognosis [231–233]. However, CRP would seem to be involved in the neuropsychiatric symptoms related to COVID-19 infection. In fact, some studies reported a significant association between peripheral inflammatory biomarkers and mental conditions among patients affected with COVID-19, particularly in depressive patients who have been positively correlated with elevated CRP levels [227, 234], even though another study did not report any association between CRP and post-COVID depression at the third month of follow-up [235]. Every 50 mg/L CRP increase has been associated with a higher risk of delirium onset in a sample of patients aged 65 years and older affected with COVID-19 [236]. Further studies reported a significant correlation between CRP levels and long-term cognitive impairment [237], with poor verbal fluency and executive function [238], as well as poor sustained attention [239], in COVID-19 patients. Therefore, this preliminary evidence may help in guiding further research studies for investigating whether CRP may be a biomarker of post-COVID depression.

## 7.12 Conclusion

Although several studies investigated the role of inflammatory biomarkers in peripheral blood in psychiatric conditions, it is still unclear whether there is a causal association between an inflammatory dysregulation and the development of a psychiatric disorder. However, measuring peripheral CRP as part of routine clinical assessment could be highly useful, because it would allow to identify a subgroup of patients in whom there is a low-grade inflammatory state, which has been demonstrated in some studies to be significantly correlated with a more severe, earlier onset, a different clinical course and prognosis and a higher percentage of treatment resistance of some mental illnesses. However, as these preliminary findings mainly come from extremely heterogeneous methodological studies with often small sample size and missing a control group, further larger longitudinal and randomized controlled studies should be implemented before drawing up clear and definitive clinical and therapeutic strategies. Overall, one could argue that clearly identifying a sensitive and specific biomarker (e.g. CRP) may potentially help

clinicians in identifying a subset of psychiatric patients who are more likely to benefit from anti-inflammatory and/or immunomodulatory treatments.

## References


1. Marnell L, Mold C, Du Clos TW. C-reactive protein: ligands, receptors and role in inflammation. Clin Immunol. 2005;117(2):104–11. https://doi.org/10.1016/j.clim.2005.08.004.
2. Sproston NR, Ashworth JJ. Role of C-reactive protein at sites of inflammation and infection. Front Immunol. 2018;9:754. https://doi.org/10.3389/fimmu.2018.00754.
3. Nehring SM, Goyal A, Bansal P, Patel BC. C reactive protein. Treasure Island, FL: StatPearls; 2021. https://www.ncbi.nlm.nih.gov/books/NBK441843/.
4. Tillett WS, Francis T. Serological reactions in pneumonia with a non-protein somatic fraction of pneumococcus. J Exp Med. 1930;52(4):561–71. https://doi.org/10.1084/jem.52.4.561.
5. Wu Y, Potempa LA, El Kebir D, Filep JG. C-reactive protein and inflammation: conformational changes affect function. Biol Chem. 2015;396(11):1181–97. https://doi.org/10.1515/hsz-2015-0149.
6. Thiele JR, Habersberger J, Braig D, Schmidt Y, Goerendt K, Maurer V, et al. Dissociation of pentameric to monomeric C-reactive protein localizes and aggravates inflammation: in vivo proof of a powerful proinflammatory mechanism and a new anti-inflammatory strategy. Circulation. 2014;130(1):35–50. https://doi.org/10.1161/CIRCULATIONAHA.113.007124.
7. Joseph J, Depp C, Martin AS, Daly RE, Glorioso DK, Palmer BW, et al. Associations of high sensitivity C-reactive protein levels in schizophrenia and comparison groups. Schizophr Res ottobre. 2015;168(1–2):456–60. https://doi.org/10.1016/j.schres.2015.08.019.
8. Ridker PM. Clinical application of C-reactive protein for cardiovascular disease detection and prevention. Circulation. 2003;107(3):363–9. https://doi.org/10.1161/01.cir.0000053730.47739.3c.
9. Orsolini L, Sarchione F, Vellante F, Fornaro M, Matarazzo I, Martinotti G, et al. Protein-C reactive as biomarker predictor of schizophrenia phases of illness?: a systematic review. Curr Neuropharmacol. 2018;16(5):583–606. https://doi.org/10.2174/1570159X16666180119144538.
10. Pearson TA, Mensah GA, Alexander RW, Anderson JL, Cannon RO, Criqui M, et al. Markers of inflammation and cardiovascular disease: application to clinical and public health practice: a statement for healthcare professionals from the Centers for Disease Control and Prevention and the American Heart Association. Circulation. 2003;107(3):499–511. https://doi.org/10.1161/01.cir.0000052939.59093.45.
11. Moylan S, Berk M, Dean OM, Samuni Y, Williams LJ, O'Neil A, et al. Oxidative & nitrosative stress in depression: why so much stress? Neurosci Biobehav Rev. 2014;45:46–62. https://doi.org/10.1016/j.neubiorev.2014.05.007.
12. Michopoulos V, Powers A, Gillespie CF, Ressler KJ, Jovanovic T. Inflammation in fear- and anxiety-based disorders: PTSD, GAD, and beyond. Neuropsychopharmacology. 2017;42(1):254–70. https://doi.org/10.1038/npp.2016.146.
13. Rosenblat JD, McIntyre RS. Bipolar disorder and immune dysfunction: epidemiological findings, proposed pathophysiology and clinical implications. Brain Sci. 2017;7(11):E144.
14. Dubois T, Reynaert C, Jacques D, Lepiece B, Patigny P, Zdanowicz N. Immunity and psychiatric disorders: variabilities of immunity biomarkers are they specific? Psychiatr Danub. 2018;30(Suppl 7):447–51.
15. Horn SR, Long MM, Nelson BW, Allen NB, Fisher PA, Byrne ML. Replication and reproducibility issues in the relationship between C-reactive protein and depression: a systematic review and focused meta-analysis. Brain Behav Immun. 2018;73:85–114. https://doi.org/10.1016/j.bbi.2018.06.016.

16. Renna ME, O'Toole MS, Spaeth PE, Lekander M, Mennin DS. The association between anxiety, traumatic stress, and obsessive-compulsive disorders and chronic inflammation: a systematic review and meta-analysis. Depress Anxiety. 2018;35(11):1081–94. https://doi.org/10.1002/da.22790.
17. Felger JC, Haroon E, Patel TA, Goldsmith DR, Wommack EC, Woolwine BJ, et al. What does plasma CRP tell us about peripheral and central inflammation in depression? Mol Psychiatry. 2020;25(6):1301–11. https://doi.org/10.1038/s41380-018-0096-3.
18. Sulhan S, Lyon KA, Shapiro LA, Huang JH. Neuroinflammation and blood-brain barrier disruption following traumatic brain injury: pathophysiology and potential therapeutic targets. J Neurosci Res. 2020;98(1):19–28. https://doi.org/10.1002/jnr.24331.
19. Turna J, Grosman Kaplan K, Anglin R, Patterson B, Soreni N, Bercik P, et al. The gut microbiome and inflammation in obsessive-compulsive disorder patients compared to age- and sex-matched controls: a pilot study. Acta Psychiatr Scand. 2020;142(4):337–47. https://doi.org/10.1111/acps.13175.
20. Caldirola D, Daccò S, Cuniberti F, Grassi M, Lorusso S, Diaferia G, et al. Elevated C-reactive protein levels across diagnoses: the first comparison among inpatients with major depressive disorder, bipolar disorder, or obsessive–compulsive disorder. J Psychosom Res. 2021;150:110604. https://doi.org/10.1016/j.jpsychores.2021.110604.
21. Woo JJ, Pouget JG, Zai CC, Kennedy JL. The complement system in schizophrenia: where are we now and what's next? Mol Psychiatry. 2020;25(1):114–30. https://doi.org/10.1038/s41380-019-0479-0.
22. D'Mello C, Le T, Swain MG. Cerebral microglia recruit monocytes into the brain in response to tumor necrosis factor signaling during peripheral organ inflammation. J Neurosci. 2009;29(7):2089–102. https://doi.org/10.1523/JNEUROSCI.3567-08.2009.
23. McKim DB, Weber MD, Niraula A, Sawicki CM, Liu X, Jarrett BL, et al. Microglial recruitment of IL-1β-producing monocytes to brain endothelium causes stress-induced anxiety. Mol Psychiatry. 2018;23(6):1421–31. https://doi.org/10.1038/mp.2017.64.
24. Wesselingh R, Butzkueven H, Buzzard K, Tarlinton D, O'Brien TJ, Monif M. Innate immunity in the central nervous system: a missing piece of the autoimmune encephalitis puzzle? Front Immunol. 2019;10:2066. https://doi.org/10.3389/fimmu.2019.02066.
25. Aveleira CA, Lin C-M, Abcouwer SF, Ambrosio AF, Antonetti DA. TNF- signals through PKC/NF- B to alter the tight junction complex and increase retinal endothelial cell permeability. Diabetes. 2010;59(11):2872–82. https://doi.org/10.2337/db09-1606.
26. Menard C, Pfau ML, Hodes GE, Kana V, Wang VX, Bouchard S, et al. Social stress induces neurovascular pathology promoting depression. Nat Neurosci. 2017;20(12):1752–60. https://doi.org/10.1016/j.bbi.2016.09.008.
27. Jeon M-T, Kim K-S, Kim ES, Lee S, Kim J, Hoe H-S, et al. Emerging pathogenic role of peripheral blood factors following BBB disruption in neurodegenerative disease. Ageing Res Rev. 2021;68:101333. https://doi.org/10.1016/j.arr.2021.101333.
28. Prakash R, Carmichael ST. Blood–brain barrier breakdown and neovascularization processes after stroke and traumatic brain injury. Curr Opin Neurol. 2015;28(6):556–64. https://doi.org/10.1097/WCO.0000000000000248.
29. Kuhlmann CRW, Librizzi L, Closhen D, Pflanzner T, Lessmann V, Pietrzik CU, et al. Mechanisms of C-reactive protein-induced blood–brain barrier disruption. Stroke. 2009;40(4):1458–66. https://doi.org/10.1161/STROKEAHA.108.535930.
30. Lim GY, Tam WW, Lu Y, Ho CS, Zhang MW, Ho RC. Prevalence of depression in the community from 30 countries between 1994 and 2014. Sci Rep. 2018;8(1):2861. https://doi.org/10.1038/s41598-018-21243-x.
31. Perez-Caballero L, Torres-Sanchez S, Romero-López-Alberca C, González-Saiz F, Mico JA, Berrocoso E. Monoaminergic system and depression. Cell Tissue Res. 2019;377(1):107–13. https://doi.org/10.1007/s00441-018-2978-8.
32. Maes M, Yirmyia R, Noraberg J, Brene S, Hibbeln J, Perini G, et al. The inflammatory & neurodegenerative (I&ND) hypothesis of depression: leads for future research and new drug

developments in depression. Metab Brain Dis. 2009;24(1):27–53. https://doi.org/10.1007/s11011-008-9118-1.
33. Keller J, Gomez R, Williams G, Lembke A, Lazzeroni L, Murphy GM, et al. HPA axis in major depression: cortisol, clinical symptomatology and genetic variation predict cognition. Mol Psychiatry. 2017;22(4):527–36. https://doi.org/10.1038/mp.2016.120.
34. Czarny P, Wigner P, Galecki P, Sliwinski T. The interplay between inflammation, oxidative stress, DNA damage, DNA repair and mitochondrial dysfunction in depression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2018;80:309–21. https://doi.org/10.1016/j.pnpbp.2017.06.036.
35. Gałecki P, Talarowska M. Inflammatory theory of depression. Psychiatr Pol. 2018;52(3):437–47.
36. Uchida S, Yamagata H, Seki T, Watanabe Y. Epigenetic mechanisms of major depression: targeting neuronal plasticity. Psychiatry Clin Neurosci. 2018;72(4):212–27. https://doi.org/10.1111/pcn.12621.
37. Gałecki P, Talarowska M. Neurodevelopmental theory of depression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2018;80:267–72. https://doi.org/10.1016/j.pnpbp.2017.05.023.
38. Osimo EF, Pillinger T, Rodriguez IM, Khandaker GM, Pariante CM, Howes OD. Inflammatory markers in depression: a meta-analysis of mean differences and variability in 5,166 patients and 5,083 controls. Brain Behav Immun. 2020;87:901–9. https://doi.org/10.1016/j.bbi.2020.02.010.
39. Dowlati Y, Herrmann N, Swardfager W, Liu H, Sham L, Reim EK, et al. A meta-analysis of cytokines in major depression. Biol Psychiatry. 2010;67(5):446–57. https://doi.org/10.1016/j.biopsych.2009.09.033.
40. Haapakoski R, Mathieu J, Ebmeier KP, Alenius H, Kivimäki M. Cumulative meta-analysis of interleukins 6 and 1β, tumour necrosis factor α and C-reactive protein in patients with major depressive disorder. Brain Behav Immun. 2015;49:206–15. https://doi.org/10.1016/j.bbi.2015.06.001.
41. Goldsmith DR, Rapaport MH, Miller BJ. A meta-analysis of blood cytokine network alterations in psychiatric patients: comparisons between schizophrenia, bipolar disorder and depression. Mol Psychiatry. 2016;21(12):1696–709. https://doi.org/10.1038/mp.2016.3.
42. Nobis A, Zalewski D, Waszkiewicz N. Peripheral markers of depression. J Clin Med. 2020;9(12):3793. https://doi.org/10.3390/jcm9123793.
43. Dantzer R. Cytokine, sickness behavior, and depression. Neurol Clin. 2006;24(3):441–60. https://doi.org/10.1016/j.ncl.2006.03.003.
44. Wium-Andersen MK, Ørsted DD, Nielsen SF, Nordestgaard BG. Elevated C-reactive protein levels, psychological distress, and depression in 73 131 individuals. JAMA Psychiat. 2013;70(2):176. https://doi.org/10.1001/2013.jamapsychiatry.102.
45. Bjerkeset O, Romild U, Smith GD, Hveem K. The associations of high levels of C-reactive protein with depression and myocardial infarction in 9258 women and men from the HUNT population study. Psychol Med. 2011;41(2):345–52. https://doi.org/10.1017/S0033291710000887.
46. Pasco JA, Nicholson GC, Williams LJ, Jacka FN, Henry MJ, Kotowicz MA, et al. Association of high-sensitivity C-reactive protein with de novo major depression. Br J Psychiatry. 2010;197(5):372–7. https://doi.org/10.1192/bjp.bp.109.076430.
47. Hickman RJ, Khambaty T, Stewart JC. C-reactive protein is elevated in atypical but not nonatypical depression: data from the National Health and nutrition examination survey (NHANES) 1999–2004. J Behav Med. 2014;37(4):621–9. https://doi.org/10.1007/s10865-013-9510-0.
48. Duivis HE, Vogelzangs N, Kupper N, de Jonge P, Penninx BWJH. Differential association of somatic and cognitive symptoms of depression and anxiety with inflammation: findings from the Netherlands study of depression and anxiety (NESDA). Psychoneuroendocrinology. 2013;38(9):1573–85. https://doi.org/10.1016/j.psyneuen.2013.01.002.

49. Zalli A, Jovanova O, Hoogendijk WJG, Tiemeier H, Carvalho LA. Low-grade inflammation predicts persistence of depressive symptoms. Psychopharmacology. 2016;233(9):1669–78. https://doi.org/10.1007/s00213-015-3919-9.
50. Gimeno D, Kivimäki M, Brunner EJ, Elovainio M, De Vogli R, Steptoe A, et al. Associations of C-reactive protein and interleukin-6 with cognitive symptoms of depression: 12-year follow-up of the Whitehall II study. Psychol Med. 2009;39(3):413–23. https://doi.org/10.1017/S0033291708003723.
51. Köhler-Forsberg O, Buttenschøn HN, Tansey KE, Maier W, Hauser J, Dernovsek MZ, et al. Association between C-reactive protein (CRP) with depression symptom severity and specific depressive symptoms in major depression. Brain Behav Immun. 2017;62:344–50. https://doi.org/10.1016/j.bbi.2017.02.020.
52. Chamberlain SR, Cavanagh J, de Boer P, Mondelli V, Jones DNC, Drevets WC, et al. Treatment-resistant depression and peripheral C-reactive protein. Br J Psychiatry. 2019;214(1):11–9. https://doi.org/10.1192/bjp.2018.66.
53. Mocking RJT, Nap TS, Westerink AM, Assies J, Vaz FM, Koeter MWJ, et al. Biological profiling of prospective antidepressant response in major depressive disorder: associations with (neuro)inflammation, fatty acid metabolism, and amygdala-reactivity. Psychoneuroendocrinology. 2017;79:84–92. https://doi.org/10.1016/j.psyneuen.2017.02.019.
54. Uher R, Tansey KE, Dew T, Maier W, Mors O, Hauser J, et al. An inflammatory biomarker as a differential predictor of outcome of depression treatment with Escitalopram and Nortriptyline. Am J Psychiatry. 2014;171(12):1278–86. https://doi.org/10.1176/appi.ajp.2014.14010094.
55. Khandaker GM, Pearson RM, Zammit S, Lewis G, Jones PB. Association of Serum Interleukin 6 and C-reactive protein in childhood with depression and psychosis in young adult life: a population-based longitudinal study. JAMA Psychiat. 2014;71(10):1121. https://doi.org/10.1001/jamapsychiatry.2014.1332.
56. Osimo EF, Baxter LJ, Lewis G, Jones PB, Khandaker GM. Prevalence of low-grade inflammation in depression: a systematic review and meta-analysis of CRP levels. Psychol Med. 2019;49(12):1958–70. https://doi.org/10.1017/S0033291719001454.
57. O'Brien SM, Scully P, Fitzgerald P, Scott LV, Dinan TG. Plasma cytokine profiles in depressed patients who fail to respond to selective serotonin reuptake inhibitor therapy. J Psychiatr Res. 2007;41(3–4):326–31. https://doi.org/10.1016/j.jpsychires.2006.05.013.
58. Chang HH, Lee IH, Gean PW, Lee S-Y, Chi MH, Yang YK, et al. Treatment response and cognitive impairment in major depression: association with C-reactive protein. Brain Behav Immun. 2012;26(1):90–5. https://doi.org/10.1016/j.bbi.2011.07.239.
59. Tuglu C, Kara SH, Caliyurt O, Vardar E, Abay E. Increased serum tumor necrosis factor-alpha levels and treatment response in major depressive disorder. Psychopharmacology. 2003;170(4):429–33. https://doi.org/10.1007/s00213-003-1566-z.
60. Köhler CA, Freitas TH, Stubbs B, Maes M, Solmi M, Veronese N, et al. Peripheral alterations in cytokine and chemokine levels after antidepressant drug treatment for major depressive disorder: systematic review and meta-analysis. Mol Neurobiol. 2017;55(5):4195–206. https://doi.org/10.1007/s12035-017-0632-1.
61. Mosiołek A, Pięta A, Jakima S, Zborowska N, Mosiołek J, Szulc A. Effects of antidepressant treatment on peripheral biomarkers in patients with major depressive disorder (MDD). J Clin Med. 2021;10(8):1706. https://doi.org/10.3390/jcm10081706.
62. Raison CL, Rutherford RE, Woolwine BJ, Shuo C, Schettler P, Drake DF, et al. A randomized controlled trial of the tumor necrosis factor antagonist infliximab for treatment-resistant depression: the role of baseline inflammatory biomarkers. JAMA Psychiat. 2013;70(1):31. https://doi.org/10.1001/2013.jamapsychiatry.4.
63. Köhler O, Benros ME, Nordentoft M, Farkouh ME, Iyengar RL, Mors O, et al. Effect of anti-inflammatory treatment on depression, depressive symptoms, and adverse effects: a systematic review and meta-analysis of randomized clinical trials. JAMA Psychiat. 2014;71(12):1381. https://doi.org/10.1001/jamapsychiatry.2014.1611.

64. Eyre HA, Baune BT. Anti-inflammatory intervention in depression. JAMA Psychiatry. 2015;72(5):511. https://doi.org/10.1001/jamapsychiatry.2014.3128.
65. Eyre HA, Air T, Proctor S, Rositano S, Baune BT. A critical review of the efficacy of non-steroidal anti-inflammatory drugs in depression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2015;57:11–6. https://doi.org/10.1016/j.pnpbp.2014.10.003.
66. Kappelmann N, Lewis G, Dantzer R, Jones PB, Khandaker GM. Antidepressant activity of anti-cytokine treatment: a systematic review and meta-analysis of clinical trials of chronic inflammatory conditions. Mol Psychiatry. 2018;23(2):335–43. https://doi.org/10.1038/mp.2016.167.
67. Bekhbat M, Chu K, Le N-A, Woolwine BJ, Haroon E, Miller AH, et al. Glucose and lipid-related biomarkers and the antidepressant response to infliximab in patients with treatment-resistant depression. Psychoneuroendocrinology. 2018;98:222–9. https://doi.org/10.1016/j.psyneuen.2018.09.004.
68. Bavaresco DV, Uggioni MLR, Ferraz SD, Marques RMM, Simon CS, Dagostin VS, et al. Efficacy of infliximab in treatment-resistant depression: a systematic review and meta-analysis. Pharmacol Biochem Behav. 2020;188:172838. https://doi.org/10.1016/j.pbb.2019.172838.
69. Liu H, Zhang Y, Gao Y, Zhang Z. Elevated levels of Hs-CRP and IL-6 after delivery are associated with depression during the 6 months post partum. Psychiatry Res. 2016;243:43–8. https://doi.org/10.1016/j.psychres.2016.02.022.
70. Buglione-Corbett R, Deligiannidis K, Leung K, Zhang N, Lee M, Rosal M, et al. Expression of inflammatory markers in women with perinatal depressive symptoms. Arch Womens Ment Health. 2018;21(6):671–9. https://doi.org/10.1007/s00737-018-0834-1.
71. Lambert M, Gressier F. Biomarqueurs de L'inflammation et dépression du post-partum: une revue systématique de la littérature. Can J Psychiatr. 2019;64(7):471–81. https://doi.org/10.1177/0706743719828970.
72. Miller ES, Hoxha D, Pinheiro E, Grobman WA, Wisner KL. The association of serum C-reactive protein with the occurrence and course of postpartum depression. Arch Womens Ment Health. 2019;22(1):129–32.
73. Roomruangwong C, Kanchanatawan B, Sirivichayakul S, Mahieu B, Nowak G, Maes M. Lower serum zinc and higher CRP strongly predict prenatal depression and Physio-somatic symptoms, which all together predict postnatal depressive symptoms. Mol Neurobiol. 2017;54(2):1500–12. https://doi.org/10.1007/s12035-016-9741-5.
74. Aas M, Vecchio C, Pauls A, Mehta M, Williams S, Hazelgrove K, et al. Biological stress response in women at risk of postpartum psychosis: the role of life events and inflammation. Psychoneuroendocrinology. 2020;113:104558. https://doi.org/10.1016/j.psyneuen.2019.104558.
75. Morgan JE, Lee SS, Mahrer NE, Guardino CM, Davis EP, Shalowitz MU, et al. Prenatal maternal C-reactive protein prospectively predicts child executive functioning at ages 4–6 years. Dev Psychobiol. 2020;62(8):1111–23. https://doi.org/10.1002/dev.21982.
76. Hunter SK, Hoffman MC, D'Alessandro A, Noonan K, Wyrwa A, Freedman R, et al. Male fetus susceptibility to maternal inflammation: C-reactive protein and brain development. Psychol Med. 2021;51(3):450–9. https://doi.org/10.1017/S0033291719003313.
77. Shao S, Wang J, Huang K, Wang S, Liu H, Wan S, et al. Prenatal pregnancy-related anxiety predicts boys' ADHD symptoms via placental C-reactive protein. Psychoneuroendocrinology. 2020;120:104797. https://doi.org/10.1016/j.psyneuen.2020.104797.
78. Canetta S, Sourander A, Surcel H-M, Hinkka-Yli-Salomäki S, Leiviskä J, Kellendonk C, et al. Elevated maternal C-reactive protein and increased risk of schizophrenia in a national birth cohort. Am J Psychiatry. 2014;171(9):960–8. https://doi.org/10.1176/appi.ajp.2014.13121579.
79. Brown AS, Sourander A, Hinkka-Yli-Salomäki S, McKeague IW, Sundvall J, Surcel H-M. Elevated maternal C-reactive protein and autism in a national birth cohort. Mol Psychiatry. 2014;19(2):259–64. https://doi.org/10.1038/mp.2012.197.

80. Chudal R, Sourander A, Surcel H-M, Sucksdorff D, Hinkka-Yli-Salomäki S, Brown AS. Gestational maternal C—reactive protein and risk of bipolar disorder among young individuals in a Nationwide birth cohort. J Affect Disord. 2017;208:41–6. https://doi.org/10.1016/j.jad.2016.08.056.
81. Chudal R, Brown AS, Gyllenberg D, Hinkka-Yli-Salomäki S, Sucksdorff M, Surcel H-M, et al. Maternal serum C-reactive protein (CRP) and offspring attention deficit hyperactivity disorder (ADHD). Eur Child Adolesc Psychiatry. 2020;29(2):239–47. https://doi.org/10.1007/s00787-019-01372-y.
82. Carvalho AF, Firth J, Vieta E, Ropper AH. Bipolar disorder. N Engl J Med. 2020;383(1):58–66. https://doi.org/10.1056/NEJMra1906193.
83. Marshe VS, Pira S, Mantere O, Bosche B, Looper KJ, Herrmann N, et al. C-reactive protein and cardiovascular risk in bipolar disorder patients: a systematic review. Prog Neuro-Psychopharmacol Biol Psychiatry. 2017;79:442–51. https://doi.org/10.1016/j.pnpbp.2017.07.026.
84. Nielsen RE, Banner J, Jensen SE. Cardiovascular disease in patients with severe mental illness. Nat Rev Cardiol. 2021;18(2):136–45. https://doi.org/10.1038/s41569-020-00463-7.
85. Solmi M, Suresh Sharma M, Osimo EF, Fornaro M, Bortolato B, Croatto G, et al. Peripheral levels of C-reactive protein, tumor necrosis factor-α, interleukin-6, and interleukin-1β across the mood spectrum in bipolar disorder: a meta-analysis of mean differences and variability. Brain Behav Immun. 2021;97:193–203. https://doi.org/10.1016/j.bbi.2021.07.014.
86. Pitchot W, Scantamburlo G, Ansseau M, Souery D. Le trouble bipolaire: une affection bien complexe. Rev Méd Liège. 2012;67(5–6):366–73.
87. Landgraf D, McCarthy MJ, Welsh DK. Circadian clock and stress interactions in the molecular biology of psychiatric disorders. Curr Psychiatry Rep. 2014;16(10):483. https://doi.org/10.1007/s11920-014-0483-7.
88. Sayana P, Colpo GD, Simões LR, Giridharan VV, Teixeira AL, Quevedo J, et al. A systematic review of evidence for the role of inflammatory biomarkers in bipolar patients. J Psychiatr Res. 2017;92:160–82. https://doi.org/10.1016/j.jpsychires.2017.03.018.
89. Morris G, Puri BK, Walker AJ, Maes M, Carvalho AF, Bortolasci CC, et al. Shared pathways for neuroprogression and somatoprogression in neuropsychiatric disorders. Neurosci Biobehav Rev. 2019;107:862–82. https://doi.org/10.1016/j.neubiorev.2019.09.025.
90. Munkholm K, Weikop P, Kessing LV, Vinberg M. Elevated levels of IL-6 and IL-18 in manic and hypomanic states in rapid cycling bipolar disorder patients. Brain Behav Immun. 2015;43:205–13. https://doi.org/10.1016/j.bbi.2014.09.021.
91. Dargél AA, Godin O, Kapczinski F, Kupfer DJ, Leboyer M. C-reactive protein alterations in bipolar disorder: a meta-analysis. J Clin Psychiatry. 2015;76(02):142–50. https://doi.org/10.4088/JCP.14r09007.
92. Fernandes BS, Steiner J, Molendijk ML, Dodd S, Nardin P, Gonçalves C-A, et al. C-reactive protein concentrations across the mood spectrum in bipolar disorder: a systematic review and meta-analysis. Lancet Psychiatry. 2016;3(12):1147–56. https://doi.org/10.1016/S2215-0366(16)30370-4.
93. Jacoby AS, Munkholm K, Vinberg M, Pedersen BK, Kessing LV. Cytokines, brain-derived neurotrophic factor and C-reactive protein in bipolar I disorder—results from a prospective study. J Affect Disord. 2016;197:167–74. https://doi.org/10.1016/j.jad.2016.03.040.
94. Cunha ÂB, Andreazza AC, Gomes FA, Frey BN, da Silveira LE, Gonçalves CA, et al. Investigation of serum high-sensitive C-reactive protein levels across all mood states in bipolar disorder. Eur Arch Psychiatry Clin Neurosci. 2008;258(5):300–4. https://doi.org/10.1007/s00406-007-0797-0.
95. Becking K, Boschloo L, Vogelzangs N, Haarman BCM, Riemersma-van der Lek R, Penninx BWJH, et al. The association between immune activation and manic symptoms in patients with a depressive disorder. Transl Psychiatry. 2013;3(10):e314. https://doi.org/10.1038/tp.2013.87.

96. Gorgulu Y, Uluturk MK, Palabiyik O. Comparison of serum BDNF, IL-1β, IL-6, TNF-α, CRP and leucocyte levels in unipolar mania and bipolar disorder. Acta Neuropsychiatr. 2021;33(6):317–22. https://doi.org/10.1017/neu.2021.25.
97. Chang HH, Wang T-Y, Lee IH, Lee S-Y, Chen KC, Huang S-Y, et al. C-reactive protein: a differential biomarker for major depressive disorder and bipolar II disorder. World J Biol Psychiatry. 2017;18(1):63–70. https://doi.org/10.3109/15622975.2016.1155746.
98. Wium-Andersen MK, Ørsted DD, Nordestgaard BG. Elevated C-reactive protein and late-onset bipolar disorder in 78 809 individuals from the general population. Br J Psychiatry. 2016;208(2):138–45. https://doi.org/10.1192/bjp.bp.114.150870.
99. Dickerson F, Stallings C, Origoni A, Boronow J, Yolken R. Elevated serum levels of C-reactive protein are associated with mania symptoms in outpatients with bipolar disorder. Prog Neuro-Psychopharmacol Biol Psychiatry. 2007;31(4):952–5. https://doi.org/10.1016/j.pnpbp.2007.02.018.
100. Lee S-Y, Chen S-L, Chang Y-H, Chen PS, Huang S-Y, Tzeng N-S, et al. Inflammation's association with metabolic profiles before and after a twelve-week clinical trial in drug-Naïve patients with bipolar II disorder. PLoS One. 2013;8(6):e66847.
101. Hamdi G, Ammar HB, Khelifa E, Chaaben AB, Khouadja S, Ayari F, et al. High-sensitive c-reactive protein levels in euthymic bipolar patients: case-control study. Psychiatry Q. 2021;92(2):803–11. https://doi.org/10.1007/s11126-020-09854-y.
102. Boukouaci W, Oliveira J, Etain B, Bennabi M, Mariaselvam C, Hamdani N, et al. Association between CRP genetic diversity and bipolar disorder comorbid complications. Int J Bipolar Disord. 2018;6(1):4. https://doi.org/10.1186/s40345-017-0109-1.
103. Evers A-K, Veeh J, McNeill R, Reif A, Kittel-Schneider S. C-reactive protein concentration in bipolar disorder: association with genetic variants. Int J Bipolar Disord. 2019;7(1):26. https://doi.org/10.1186/s40345-019-0162-z.
104. Misiak B, Stańczykiewicz B, Kotowicz K, Rybakowski JK, Samochowiec J, Frydecka D. Cytokines and C-reactive protein alterations with respect to cognitive impairment in schizophrenia and bipolar disorder: a systematic review. Schizophr Res. 2018;192:16–29. https://doi.org/10.1016/j.schres.2017.04.015.
105. Milton DC, Ward J, Ward E, Lyall DM, Strawbridge RJ, Smith DJ, et al. The association between C-reactive protein, mood disorder, and cognitive function in UK biobank. Eur Psychiatry. 2021;64(1):e14. https://doi.org/10.1192/j.eurpsy.2021.6.
106. Millett CE, Perez-Rodriguez M, Shanahan M, Larsen E, Yamamoto HS, Bukowski C, et al. C-reactive protein is associated with cognitive performance in a large cohort of euthymic patients with bipolar disorder. Mol Psychiatry. 2021;26(8):4096–105. https://doi.org/10.1038/s41380-019-0591-1.
107. Chiu C-T, Wang Z, Hunsberger JG, Chuang D-M. Therapeutic potential of mood stabilizers lithium and Valproic acid: beyond bipolar disorder. Sibley DR, curatore. Pharmacol Rev. 2013;65(1):105–42. https://doi.org/10.1124/pr.111.005512.
108. Raison CL, Pikalov A, Siu C, Tsai J, Koblan K, Loebel A. C-reactive protein and response to lurasidone in patients with bipolar depression. Brain Behav Immun. 2018;73:717–24. https://doi.org/10.1016/j.bbi.2018.08.009.
109. Fiedorowicz JG, Cyranowski JM, Liu Z, Swartz HA. Changes in inflammation with treatment for bipolar II depression: pilot trial data on differential effects of psychotherapy and medication. Neurol Psychiatry Brain Res. 2019;33:112–8. https://doi.org/10.1016/j.npbr.2019.07.007.
110. Raison CL, Siu C, Pikalov A, Tocco M, Loebel A. C-reactive protein and response to lurasidone treatment in children and adolescents with bipolar I depression: results from a placebo-controlled trial. Brain Behav Immun. 2020;84:269–74. https://doi.org/10.1016/j.bbi.2019.12.010.
111. McIntyre RS, Subramaniapillai M, Lee Y, Pan Z, Carmona NE, Shekotikhina M, et al. Efficacy of adjunctive infliximab vs placebo in the treatment of adults with bipolar I/II

depression: a randomized clinical trial. JAMA Psychiat. 2019;76(8):783. https://doi.org/10.1001/jamapsychiatry.2019.0779.
112. Courtet P, Jaussent I, Genty C, Dupuy AM, Guillaume S, Ducasse D, et al. Increased CRP levels may be a trait marker of suicidal attempt. Eur Neuropsychopharmacol. 2015;25(10):1824–31. https://doi.org/10.1016/j.euroneuro.2015.05.003.
113. Chen X, Pu J, Liu Y, Tian L, Chen Y, Gui S, et al. Increased C-reactive protein concentrations were associated with suicidal behavior in patients with depressive disorders: a meta-analysis. Psychiatry Res. 2020;292:113320. https://doi.org/10.1016/j.psychres.2020.113320.
114. Batty GD, Bell S, Stamatakis E, Kivimäki M. Association of Systemic Inflammation with Risk of completed suicide in the general population. JAMA Psychiat. 2016;73(9):993. https://doi.org/10.1001/jamapsychiatry.2016.1805.
115. Park RJ, Kim YH. Association between high sensitivity CRP and suicidal ideation in the Korean general population. Eur Neuropsychopharmacol. 2017;27(9):885–91. https://doi.org/10.1016/j.euroneuro.2017.06.010.
116. Graham JE, Robles TF, Kiecolt-Glaser JK, Malarkey WB, Bissell MG, Glaser R. Hostility and pain are related to inflammation in older adults. Brain Behav Immun. 2006;20(4):389–400. https://doi.org/10.1016/j.bbi.2005.11.002.
117. Coccaro EF, Lee R, Coussons-Read M. Elevated plasma inflammatory markers in individuals with intermittent explosive disorder and correlation with aggression in humans. JAMA Psychiat. 2014;71(2):158. https://doi.org/10.1001/jamapsychiatry.2013.3297.
118. Coccaro EF, Lee R, Coussons-Read M. Cerebrospinal fluid and plasma C-reactive protein and aggression in personality-disordered subjects: a pilot study. J Neural Transm. 2015;122(2):321–6. https://doi.org/10.1007/s00702-014-1263-6.
119. Suchankova P, Henningsson S, Baghaei F, Rosmond R, Holm G, Ekman A. Genetic variability within the innate immune system influences personality traits in women. Genes Brain Behav. 2009;8(2):212–7. https://doi.org/10.1111/j.1601-183X.2008.00461.x.
120. Suchankova P, Holm G, Träskman-Bendz L, Brundin L, Ekman A. The +1444C>T polymorphism in the CRP gene: a study on personality traits and suicidal behaviour. Psychiatr Genet. 2013;23(2):70–6. https://doi.org/10.1097/YPG.0b013e32835d71b6.
121. Jacomb I, Stanton C, Vasudevan R, Powell H, O'Donnell M, Lenroot R, et al. C-reactive protein: higher during acute psychotic episodes and related to cortical thickness in schizophrenia and healthy controls. Front Immunol. 2018;9:2230. https://doi.org/10.3389/fimmu.2018.02230.
122. van Os J, Kenis G, Rutten BPF. The environment and schizophrenia. Nature. 2010;468(7321):203–12. https://doi.org/10.1038/nature09563.
123. Castellani CA, Melka MG, Gui JL, O'Reilly RL, Singh SM. Integration of DNA sequence and DNA methylation changes in monozygotic twin pairs discordant for schizophrenia. Schizophr Res. 2015;169(1–3):433–40. https://doi.org/10.1016/j.schres.2015.09.021.
124. van de Leemput J, Hess JL, Glatt SJ, Tsuang MT. Genetics of schizophrenia. In: Advances in genetics. Amsterdam: Elsevier; 2016. https://doi.org/10.1016/bs.adgen.2016.08.001.
125. Müller N. Immunology of schizophrenia. Neuroimmunomodulation. 2014;21(2–3):109–16. https://doi.org/10.1159/000356538.
126. Bora E. Peripheral inflammatory and neurotrophic biomarkers of cognitive impairment in schizophrenia: a meta-analysis. Psychol Med. 2019;49(12):1971–9. https://doi.org/10.1017/S0033291719001685.
127. Upthegrove R, Khandaker GM. Cytokines, oxidative stress and cellular markers of inflammation in schizophrenia. Curr Top Behav Neurosci. 2020;44:49–66. https://doi.org/10.1007/7854_2018_88.
128. Spivak B, Radwan M, Bartur P, Mester R, Weizman A. Antinuclear autoantibodies in chronic schizophrenia. Acta Psychiatr Scand. 1995;92(4):266–9. https://doi.org/10.1111/j.1600-0447.1995.tb09581.x.

129. Potvin S, Stip E, Sepehry AA, Gendron A, Bah R, Kouassi E. Inflammatory cytokine alterations in schizophrenia: a systematic quantitative review. Biol Psychiatry. 2008;63(8):801–8. https://doi.org/10.1016/j.biopsych.2007.09.024.
130. Monteiro R, Azevedo I. Chronic inflammation in obesity and the metabolic syndrome. Mediat Inflamm. 2010;2010:1–10. https://doi.org/10.1155/2010/289645.
131. Chen S-J, Chao Y-L, Chen C-Y, Chang C-M, Wu EC-H, Wu C-S, et al. Prevalence of autoimmune diseases in in-patients with schizophrenia: nationwide population-based study. Br J Psychiatry. 2012;200(5):374–80. https://doi.org/10.1192/bjp.bp.111.092098.
132. Benros ME, Pedersen MG, Rasmussen H, Eaton WW, Nordentoft M, Mortensen PB. A Nationwide study on the risk of autoimmune diseases in individuals with a personal or a family history of schizophrenia and related psychosis. Am J Psychiatry. 2014;171(2):218–26. https://doi.org/10.1176/appi.ajp.2013.13010086.
133. Juncal-Ruiz M, Riesco-Dávila L, de la Foz VO-G, Ramírez-Bonilla M, Martínez-García O, Irure-Ventura J, et al. The effect of excess weight on circulating inflammatory cytokines in drug-naïve first-episode psychosis individuals. J Neuroinflammation. 2018;15(1):63. https://doi.org/10.1186/s12974-018-1096-6.
134. Zajkowska Z, Mondelli V. First-episode psychosis: an inflammatory state? Neuroimmunomodulation. 2014;21(2–3):102–8. https://doi.org/10.1159/000356536.
135. Perkins DO, Jeffries CD, Addington J, Bearden CE, Cadenhead KS, Cannon TD, et al. Towards a psychosis risk blood diagnostic for persons experiencing high-risk symptoms: preliminary results from the NAPLS project. Schizophr Bull. 2015;41(2):419–28. https://doi.org/10.1093/schbul/sbu099.
136. Metcalf SA, Jones PB, Nordstrom T, Timonen M, Mäki P, Miettunen J, et al. Serum C-reactive protein in adolescence and risk of schizophrenia in adulthood: a prospective birth cohort study. Brain Behav Immun. 2017;59:253–9.
137. Reponen EJ, Dieset I, Tesli M, Mørch RH, Aas M, Vedal TSJ, et al. Atherogenic lipid ratios related to myeloperoxidase and C-reactive protein levels in psychotic disorders. Front Psych. 2020;11:672. https://doi.org/10.3389/fpsyt.2020.00672.
138. Fawzi MH, Fawzi MM, Fawzi MM, Said NS. C-reactive protein serum level in drug-free male Egyptian patients with schizophrenia. Psychiatry Res. 2011;190(1):91–7. https://doi.org/10.1016/j.psychres.2011.05.010.
139. Lieburg EJ, Nolte IM, Klein HC, Knegtering H. Relation of inflammatory markers with symptoms of psychotic disorders: a large cohort study. Prog Neuro-Psychopharmacol Biol Psychiatry. 2018;86:89–94. https://doi.org/10.1016/j.pnpbp.2018.04.006.
140. BramP P, Abbasi A, Wong A, Vaez A, Nolte I, Franceschini N, et al. Investigating the causal relationship of C-reactive protein with 32 complex somatic and psychiatric outcomes: a large-scale cross-consortium Mendelian randomization study. PLoS Med. 2016;13(6):e1001976. https://doi.org/10.1371/journal.pmed.1001976.
141. Micoulaud-Franchi J-A, Faugere M, Boyer L, Fond G, Richieri R, Faget C, et al. Elevated C-reactive protein is associated with sensory gating deficit in schizophrenia. Schizophr Res. 2015;165(1):94–6. https://doi.org/10.1016/j.schres.2015.03.018.
142. Bulzacka E, Boyer L, Schürhoff F, Godin O, Berna F, Brunel L, et al. Chronic peripheral inflammation is associated with cognitive impairment in schizophrenia: results from the multicentric FACE-SZ dataset. Schizophr Bull. 2016;42(5):1290–302. https://doi.org/10.1093/schbul/sbw029.
143. Chang JP-C, Mondelli V, Satyanarayanan SK, Chiang Y-J, Chen H-T, Su K-P, et al. Cortisol, inflammatory biomarkers and neurotrophins in children and adolescents with attention deficit hyperactivity disorder (ADHD) in Taiwan. Brain Behav Immun. 2020;88:105–13. https://doi.org/10.1016/j.bbi.2020.05.017.
144. Fathian F, Løberg E-M, Gjestad R, Steen VM, Kroken RA, Jørgensen HA, et al. Associations between C-reactive protein levels and cognition during the first 6 months after acute psychosis. Acta Neuropsychiatr. 2019;31(1):36–45. https://doi.org/10.1017/neu.2018.25.

145. Miller BJ, Pikalov A, Siu CO, Tocco M, Tsai J, Harvey PD, et al. Association of C-reactive protein and metabolic risk with cognitive effects of lurasidone in patients with schizophrenia. Compr Psychiatry. 2020;102:152195. https://doi.org/10.1016/j.comppsych.2020.152195.
146. Zhang XY, Zhou DF, Cao LY, Zhang PY, Wu GY, Shen YC. Changes in serum Interleukin-2, −6, and −8 levels before and during treatment with Risperidone and haloperidol: relationship to outcome in schizophrenia. J Clin Psychiatry. 2004;65(7):940–7. https://doi.org/10.4088/jcp.v65n0710.
147. Tourjman V, Kouassi É, Koué M-È, Rocchetti M, Fortin-Fournier S, Fusar-Poli P, et al. Antipsychotics' effects on blood levels of cytokines in schizophrenia: a meta-analysis. Schizophr Res. 2013;151(1–3):43–7. https://doi.org/10.1016/j.schres.2013.10.011.
148. Fond G, Resseguier N, Schürhoff F, Godin O, Andrianarisoa M, et al. The FACE-SZ (FondaMental academic centers of expertise for schizophrenia) group. Relationships between low-grade peripheral inflammation and psychotropic drugs in schizophrenia: results from the national FACE-SZ cohort. Eur Arch Psychiatry Clin Neurosci. 2018;268(6):541–53. https://doi.org/10.1007/s00406-017-0847-1.
149. Fond G, Godin O, Boyer L, Berna F, Andrianarisoa M, et al. The FACE-SZ (FondaMental academic centers of expertise for schizophrenia) group. Chronic low-grade peripheral inflammation is associated with ultra resistant schizophrenia. Results from the FACE-SZ cohort. Eur Arch Psychiatry Clin Neurosci. 2019;269(8):985–92. https://doi.org/10.1007/s00406-018-0908-0.
150. Sommer IE, de Witte L, Begemann M, Kahn RS. Nonsteroidal anti-inflammatory drugs in schizophrenia: ready for practice or a good start? A Meta-Analysis. J Clin Psychiatry. 2012;73(04):414–9. https://doi.org/10.4088/JCP.10r06823.
151. Marini S, De Berardis D, Vellante F, Santacroce R, Orsolini L, Valchera A, et al. Celecoxib adjunctive treatment to antipsychotics in schizophrenia: a review of randomized clinical add-on trials. Mediat Inflamm. 2016;2016:1–8. https://doi.org/10.1155/2016/3476240.
152. Çakici N, van Beveren NJM, Judge-Hundal G, Koola MM, Sommer IEC. An update on the efficacy of anti-inflammatory agents for patients with schizophrenia: a meta-analysis. Psychol Med. 2019;49(14):2307–19. https://doi.org/10.1017/S0033291719001995.
153. American Psychiatric Association. Diagnostic and statistical manual of mental disorders. 5th ed. Washington, DC: American Psychiatric Association; 2013.
154. Pitsavos C, Panagiotakos DB, Papageorgiou C, Tsetsekou E, Soldatos C, Stefanadis C. Anxiety in relation to inflammation and coagulation markers, among healthy adults: the ATTICA study. Atherosclerosis. 2006;185(2):320–6. https://doi.org/10.1016/j.atherosclerosis.2005.06.001.
155. Copeland WE, Shanahan L, Worthman C, Angold A, Costello EJ. Generalized anxiety and C-reactive protein levels: a prospective, longitudinal analysis. Psychol Med. 2012;42(12):2641–50. https://doi.org/10.1017/S0033291712000554.
156. Vogelzangs N, Beekman ATF, de Jonge P, Penninx BWJH. Anxiety disorders and inflammation in a large adult cohort. Transl Psychiatry. 2013;3(4):e249. https://doi.org/10.1038/tp.2013.27.
157. Wagner E-YN, Wagner JT, Glaus J, Vandeleur CL, Castelao E, Strippoli M-PF, et al. Evidence for chronic low-grade systemic inflammation in individuals with agoraphobia from a population-based prospective study. PLoS One. 2015;10(4):e0123757. https://doi.org/10.1371/journal.pone.0123757.
158. O'Donovan A, Hughes BM, Slavich GM, Lynch L, Cronin M-T, O'Farrelly C, et al. Clinical anxiety, cortisol and interleukin-6: evidence for specificity in emotion–biology relationships. Brain Behav Immun. 2010;24(7):1074–7. https://doi.org/10.1016/j.bbi.2010.03.003.
159. Costello H, Gould RL, Abrol E, Howard R. Systematic review and meta-analysis of the association between peripheral inflammatory cytokines and generalised anxiety disorder. BMJ Open. 2019;9(7):e027925. https://doi.org/10.1136/bmjopen-2018-027925.
160. De Berardis D, Serroni N, Campanella D, Marini S, Rapini G, Valchera A, et al. Alexithymia, suicide ideation, C-reactive protein, and serum lipid levels among outpatients with generalized

anxiety disorder. Arch Suicide Res. 2017;21(1):100–12. https://doi.org/10.1080/13811118.2015.1004485.
161. Khandaker GM, Zammit S, Lewis G, Jones PB. Association between serum C-reactive protein and DSM-IV generalized anxiety disorder in adolescence: findings from the ALSPAC cohort. Neurobiol Stress. 2016;4:55–61. https://doi.org/10.1016/j.bbi.2017.11.020.
162. Nayek S, Ghosh S. A comparative study of serum C-reactive protein in patients with generalised anxiety disorder and depression. J Med Res. 2018;4:123–31.
163. Tang Z, Ye G, Chen X, Pan M, Fu J, Fu T, et al. Peripheral proinflammatory cytokines in Chinese patients with generalised anxiety disorder. J Affect Disord. 2018;225:593–8. https://doi.org/10.1016/j.jad.2017.08.082.
164. Korkeila J, Runsten S, Ollikainen S, Korkeila K. PW01-161—generalized anxiety disorder and immunity markers in a stratified population sample. Eur Psychiatry. 2010;25(S1):1–1.
165. Naudé PJW, Roest AM, Stein DJ, de Jonge P, Doornbos B. Anxiety disorders and CRP in a population cohort study with 54,326 participants: the life lines study. World J Biol Psychiatry. 2018;19(6):461–70. https://doi.org/10.1080/15622975.2018.1433325.
166. Friend SF, Nachnani R, Powell SB, Risbrough VB. C-reactive protein: marker of risk for posttraumatic stress disorder and its potential for a mechanistic role in trauma response and recovery. Eur J Neurosci. 2020;55(9-10):2297–310. https://doi.org/10.1111/ejn.15031.
167. Afzali B, Lombardi G, Lechler RI, Lord GM. The role of T helper 17 (Th17) and regulatory T cells (Treg) in Human organ transplantation and autoimmune disease: role of Th17 and Tregs in Human disease. Clin Exp Immunol. 2007;148(1):32–46. https://doi.org/10.1111/j.1365-2249.2007.03356.x.
168. Sommershof A, Aichinger H, Engler H, Adenauer H, Catani C, Boneberg E-M, et al. Substantial reduction of naïve and regulatory T cells following traumatic stress. Brain Behav Immun. 2009;23(8):1117–24. https://doi.org/10.1016/j.bbi.2009.07.003.
169. Baumeister D, Akhtar R, Ciufolini S, Pariante CM, Mondelli V. Childhood trauma and adulthood inflammation: a meta-analysis of peripheral C-reactive protein, interleukin-6 and tumour necrosis factor-α. Mol Psychiatry. 2016;21(5):642–9. https://doi.org/10.1038/mp.2015.67.
170. Fischer KF, Simon MS, Elsner J, Dobmeier J, Dorr J, Blei L, et al. Assessing the links between childhood trauma, C-reactive protein and response to antidepressant treatment in patients with affective disorders. Eur Arch Psychiatry Clin Neurosci. 2021;271(7):1331–41. https://doi.org/10.1007/s00406-021-01245-z.
171. Lacey RE, Kumari M, McMunn A. Parental separation in childhood and adult inflammation: the importance of material and psychosocial pathways. Psychoneuroendocrinology. 2013;38(11):2476–84. https://doi.org/10.1016/j.psyneuen.2013.05.007.
172. McDade TW, Hoke M, Borja JB, Adair LS, Kuzawa C. Do environments in infancy moderate the association between stress and inflammation in adulthood? Initial evidence from a birth cohort in the Philippines. Brain Behav Immun. 2013;31:23–30. https://doi.org/10.1016/j.bbi.2012.08.010.
173. Powers A, Dixon HD, Conneely K, Gluck R, Munoz A, Rochat C, et al. The differential effects of PTSD, MDD, and dissociation on CRP in trauma-exposed women. Compr Psychiatry. 2019;93:33–40. https://doi.org/10.1016/j.comppsych.2019.06.007.
174. Söndergaard HP, Hansson L-O, Theorell T. The inflammatory markers C-reactive protein and serum amyloid a in refugees with and without posttraumatic stress disorder. Clin Chim Acta. 2004;342(1–2):93–8. https://doi.org/10.1016/j.cccn.2003.12.019.
175. McCanlies EC, Araia SK, Joseph PN, Mnatsakanova A, Andrew ME, Burchfiel CM, et al. C-reactive protein, Interleukin-6, and posttraumatic stress disorder symptomology in urban police officers. Cytokine. 2011;55(1):74–8. https://doi.org/10.1016/j.cyto.2011.03.025.
176. Heath NM, Chesney SA, Gerhart JI, Goldsmith RE, Luborsky JL, Stevens NR, et al. Interpersonal violence, PTSD, and inflammation: potential psychogenic pathways to higher C-reactive protein levels. Cytokine. 2013;63(2):172–8. https://doi.org/10.1016/j.cyto.2013.04.030.

177. Eraly SA, Nievergelt CM, Maihofer AX, Barkauskas DA, Biswas N, Agorastos A, et al. Assessment of plasma C-reactive protein as a biomarker of posttraumatic stress disorder risk. JAMA Psychiat. 2014;71(4):423. https://doi.org/10.1001/jamapsychiatry.2013.4374.
178. Passos IC, Vasconcelos-Moreno MP, Costa LG, Kunz M, Brietzke E, Quevedo J, et al. Inflammatory markers in post-traumatic stress disorder: a systematic review, meta-analysis, and meta-regression. Lancet Psychiatry. 2015;2(11):1002–12.
179. Rosen RL, Levy-Carrick N, Reibman J, Xu N, Shao Y, Liu M, et al. Elevated C-reactive protein and posttraumatic stress pathology among survivors of the 9/11 world trade center attacks. J Psychiatr Res. 2017;89:14–21.
180. Yang J-J, Jiang W. Immune biomarkers alterations in post-traumatic stress disorder: a systematic review and meta-analysis. J Affect Disord. 2020;268:39–46. https://doi.org/10.1016/j.jad.2020.02.044.
181. Miller RJ, Sutherland AG, Hutchison JD, Alexander DA. C-reactive protein and interleukin6 receptor in post-traumatic stress disorder: a pilot study. Cytokine. 2001;13(4):253–5. https://doi.org/10.1006/cyto.2000.0825.
182. Sumner JA, Chen Q, Roberts AL, Winning A, Rimm EB, Gilsanz P, et al. Posttraumatic stress disorder onset and inflammatory and endothelial function biomarkers in women. Brain Behav Immun. 2018;69:203–9. https://doi.org/10.1016/j.bbi.2017.11.013.
183. Wolf EJ, Logue MW, Zhao X, Daskalakis NP, Morrison FG, Escarfulleri S, et al. PTSD and the klotho longevity gene: evaluation of longitudinal effects on inflammation via DNA methylation. Psychoneuroendocrinology. 2020;117:104656. https://doi.org/10.1016/j.psyneuen.2020.104656.
184. Michopoulos V, Rothbaum AO, Jovanovic T, Almli LM, Bradley B, Rothbaum BO, et al. Association of *CRP* genetic variation and CRP level with elevated PTSD symptoms and physiological responses in a civilian population with high levels of trauma. Am J Psychiatry. 2015;172(4):353–62. https://doi.org/10.1176/appi.ajp.2014.14020263.
185. Miller MW, Maniates H, Wolf EJ, Logue MW, Schichman SA, Stone A, et al. CRP polymorphisms and DNA methylation of the AIM2 gene influence associations between trauma exposure, PTSD, and C-reactive protein. Brain Behav Immun. 2018;67:194–202.
186. Swedo SE, Leonard HL, Garvey M, Mittleman B, Allen AJ, Perlmutter S, et al. Pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections: clinical description of the first 50 cases. Am J Psychiatry. 1998;155(2):264–71.
187. Danışman Sonkurt M, Altınöz AE, Köşger F, Yiğitaslan S, Güleç G, Eşsizoğlu A. Are there differences in oxidative stress and inflammatory processes between the autogenous and reactive subtypes of obsessive-compulsive disorder? A controlled cross-sectional study. Braz J Psychiatry. 2021;44:S1516. https://doi.org/10.1590/1516-4446-2021-1740.
188. Mataix-Cols D, Frans E, Pérez-Vigil A, Kuja-Halkola R, Gromark C, Isomura K, et al. A total-population multigenerational family clustering study of autoimmune diseases in obsessive-compulsive disorder and Tourette's/chronic tic disorders. Mol Psychiatry. 2018;23(7):1652–8.
189. Gerentes M, Pelissolo A, Rajagopal K, Tamouza R, Hamdani N. Obsessive-compulsive disorder: autoimmunity and Neuroinflammation. Curr Psychiatry Rep. 2019;21(8):78. https://doi.org/10.1007/s11920-019-1062-8.
190. Cosco TD, Pillinger T, Emam H, Solmi M, Budhdeo S, Matthew Prina A, et al. Immune aberrations in obsessive-compulsive disorder: a systematic review and meta-analysis. Mol Neurobiol. 2019;56(7):4751–9. https://doi.org/10.1007/s12035-018-1409-x.
191. Ekinci A, Ekinci O. The relationships between low grade inflammation, demographic and clinical characteristics in patients with obsessive compulsive disorder. Anadolu Psikiyatr Derg. 2017;18(5):438. https://doi.org/10.5455/apd.256532.
192. Anney RJL, Lasky-Su J, Ó'Dúshláine C, Kenny E, Neale BM, Mulligan A, et al. Conduct disorder and ADHD: evaluation of conduct problems as a categorical and quantitative trait in the international multicentre ADHD genetics study. Am J Med Genet B Neuropsychiatr Genet. 2008;147B(8):1369–78. https://doi.org/10.1002/ajmg.b.30871.

193. Namjoo I, Alavi Naeini A, Najafi M, Aghaye Ghazvini MR, Hasanzadeh A. The relationship between antioxidants and inflammation in children with attention deficit hyperactivity disorder. Basic Clin Neurosci J. 2020;11:313–22. https://doi.org/10.32598/bcn.11.2.1489.1.
194. Banerjee TD, Middleton F, Faraone SV. Environmental risk factors for attention-deficit hyperactivity disorder. Acta Paediatr. 2007;96(9):1269–74.
195. Joseph N, Zhang-James Y, Perl A, Faraone SV. Oxidative stress and ADHD: a meta-analysis. J Atten Disord. 2015;19(11):915–24. https://doi.org/10.1177/1087054713510354.
196. Donfrancesco R, Nativio P, Borrelli E, Giua E, Andriola E, Villa MP, et al. Serum cytokines in pediatric neuropsychiatric syndromes: focus on attention deficit hyperactivity disorder. Minerva Pediatr. 2021;73(5):398–404.
197. Anand D, Colpo GD, Zeni G, Zeni CP, Teixeira AL. Attention-deficit/hyperactivity disorder and inflammation: what does current knowledge tell us? A systematic review. Front Psychiatry. 2017;8:228. https://doi.org/10.3389/fpsyt.2017.00228.
198. Cortese S, Angriman M, Comencini E, Vincenzi B, Maffeis C. Association between inflammatory cytokines and ADHD symptoms in children and adolescents with obesity: a pilot study. Psychiatry Res. 2019;278:7–11. https://doi.org/10.1016/j.psychres.2019.05.030.
199. Darwish AH, Elgohary TM, Nosair NA. Serum Interleukin-6 level in children with attention-deficit hyperactivity disorder (ADHD). J Child Neurol. 2019;34(2):61–7. https://doi.org/10.1177/0883073818809831.
200. Dunn GA, Nigg JT, Sullivan EL. Neuroinflammation as a risk factor for attention deficit hyperactivity disorder. Pharmacol Biochem Behav. 2019;182:22–34. https://doi.org/10.1016/j.pbb.2019.05.005.
201. Yang LL, Stiernborg M, Skott E, Söderström Å, Giacobini M, Lavebratt C. Proinflammatory mediators and their associations with medication and comorbid traits in children and adults with ADHD. Eur Neuropsychopharmacol. 2020;41:118–31. https://doi.org/10.1016/j.euroneuro.2020.10.005.
202. Hariri M, Djazayery A, Djalali M, Saedisomeolia A, Rahimi A, Abdolahian E. Effect of n-3 supplementation on hyperactivity, oxidative stress and inflammatory mediators in children with attention-deficit-hyperactivity disorder. Malays J Nutr. 2012;18(3):329–35.
203. Antshel KM, Zhang-James Y, Wagner KE, Ledesma A, Faraone SV. An update on the comorbidity of ADHD and ASD: a focus on clinical management. Expert Rev Neurother. 2016;16(3):279–93. https://doi.org/10.1586/14737175.2016.1146591.
204. Yin F, Wang H, Liu Z, Gao J. Association between peripheral blood levels of C-reactive protein and autism Spectrum disorder in children: a systematic review and meta-analysis. Brain Behav Immun. 2020;88:432–41. https://doi.org/10.1016/j.bbi.2020.04.008.
205. Reece AS. High-sensitivity CRP in opiate addiction: relative and age-dependent elevations. Cardiovasc Toxicol. 2012;12(2):149–57. https://doi.org/10.1007/s12012-012-9154-2.
206. Lu RB, Wang TY, Lee SY, Chen SL, Chang YH, See Chen P, et al. Correlation between interleukin-6 levels and methadone maintenance therapy outcomes. Drug Alcohol Depend. 2019;204:107516. https://doi.org/10.1016/j.drugalcdep.2019.06.018.
207. Ribeiro CB, de Oliveira Feitosa de Castro F, Dorneles GP, de Sousa Barros JB, Silva JM, Tavares C, et al. The concomitant use of cannabis and cocaine coexists with increased LPS levels and systemic inflammation in male drug users. Cytokine. 2021;141:155472. https://doi.org/10.1016/j.cyto.2021.155472.
208. Alho H. Alcohol misuse increases serum antibodies to oxidized ldl and c-reactive protein. Alcohol Alcohol. 2004;39(4):312–5. https://doi.org/10.1093/alcalc/agh059.
209. García-Calvo X, Bolao F, Sanvisens A, Zuluaga P, Tor J, Muga R, et al. Significance of markers of monocyte activation (CD163 and sCD14) and inflammation (IL-6) in patients admitted for alcohol use disorder treatment. Alcohol Clin Exp Res. 2020;44(1):152–8. https://doi.org/10.1111/acer.14228.
210. Xu YY, Ge JF, Chen J, Liang J, Pang LJ, Gao WF, et al. Evidence of a relationship between plasma Leptin, not Nesfatin-1, and craving in male alcohol-dependent patients after abstinence. Front Endocrinol. 2020;11:159. https://doi.org/10.3389/fendo.2020.00159.

211. van de Loo AJAE, Mackus M, Kwon O, Krishnakumar IM, Garssen J, Kraneveld AD, et al. The inflammatory response to alcohol consumption and its role in the pathology of alcohol hangover. J Clin Med. 2020;9(7):2081. https://doi.org/10.3390/jcm9072081.
212. Kim DJ, Kim W, Yoon SJ, Choi BM, Kim JS, Go HJ, et al. Effects of alcohol hangover on cytokine production in healthy subjects. Alcohol. 2003;31(3):167–70. https://doi.org/10.1016/j.alcohol.2003.09.003.
213. Wiese J, McPherson S, Odden MC, Shlipak MG. Effect of Opuntia ficus indica on symptoms of the alcohol hangover. Arch Intern Med. 2004;164(12):1334. https://doi.org/10.1001/archinte.164.12.1334.
214. Mammen RR, Natinga Mulakal J, Mohanan R, Maliakel B, Illathu MK. Clove bud polyphenols alleviate alterations in inflammation and oxidative stress markers associated with binge drinking: a randomized double-blinded placebo-controlled crossover study. J Med Food. 2018;21(11):1188–96. https://doi.org/10.1089/jmf.2017.4177.
215. Rajavashisth TB, Shaheen M, Norris KC, Pan D, Sinha SK, Ortega J, et al. Decreased prevalence of diabetes in marijuana users: cross-sectional data from the National Health and nutrition examination survey (NHANES) III. BMJ Open. 2012;2(1):e000494. https://doi.org/10.1136/bmjopen-2011-000494.
216. Costello EJ, Copeland WE, Shanahan L, Worthman CM, Angold A. C-reactive protein and substance use disorders in adolescence and early adulthood: a prospective analysis. Drug Alcohol Depend. 2013;133(2):712–7. https://doi.org/10.1016/j.drugalcdep.2013.08.027.
217. Alshaarawy O, Anthony JC. Cannabis smoking and serum C-reactive protein: a quantile regressions approach based on NHANES 2005–2010. Drug Alcohol Depend. 2015;147:203–7. https://doi.org/10.1016/j.drugalcdep.2014.11.017.
218. Ferguson EG, Mannes ZL, Ennis N. Is marijuana use associated with lower inflammation? Results from waves III and IV of the national longitudinal study of adolescent to adult health. Drug Alcohol Depend. 2019;198:162–7. https://doi.org/10.1016/j.drugalcdep.2019.01.021.
219. Lisano JK, Kisiolek JN, Smoak P, Phillips KT, Stewart LK. Chronic cannabis use and circulating biomarkers of neural health, stress, and inflammation in physically active individuals. Appl Physiol Nutr Metab. 2020;45(3):258–63. https://doi.org/10.1139/apnm-2019-0300.
220. Ghazavi A, Mosayebi G, Solhi H, Rafiei M, Moazzeni SM. Serum markers of inflammation and oxidative stress in chronic opium (Taryak) smokers. Immunol Lett. 2013;153(1–2):22–6. https://doi.org/10.1016/j.imlet.2013.07.001.
221. Mirzaeipour F, Azdaki N, Mohammadi GA, Addasi E. The effects of opium addiction through different administration routes on inflammatory and coagulation factors. J Kerman Univ Med Sci. 2013;20(3):292–300.
222. Azdaki N, Zardast M, Anani-Sarab G, Abdorrazaghnaejad H, Ghasemian MR, Saburi A. Comparison between Homocysteine, fibrinogen, PT, PTT, INR and CRP in male smokers with/without addiction to opium. Addict Health. 2017;9(1):17–23.
223. Perrot S, Guilbaud G, Kayser V. Effects of intraplantar morphine on paw edema and pain-related behaviour in a rat model of repeated acute inflammation. Pain. 1999;83(2):249–57. https://doi.org/10.1016/s0304-3959(99)00110-4.
224. Glattard E, Welters ID, Lavaux T, Muller AH, Laux A, Zhang D, et al. Endogenous morphine levels are increased in sepsis: a partial implication of neutrophils. PLoS One. 2010;5(1):e8791. https://doi.org/10.1371/journal.pone.0008791.
225. van Loon JP, de Grauw JC, van Dierendonck M, L'ami JJ, Back W, van Werren PR. Intra-articular opioid analgesia is effective in reducing pain and inflammation in an equine LPS induced synovitis model: analgesic and anti-inflammatory effects of intra-articular opioids in equine synovitis. Equine Vet J. 2010;42(5):412–9. https://doi.org/10.1111/j.2042-3306.2010.00077.x.
226. Alexander SA, Mathew Thomas V, Savage JA. Elevated C-reactive protein and role of steroids in cocaine-associated levamisole-induced vasculitis. Cureus. 2020;12(4):e7597. https://doi.org/10.7759/cureus.7597.

227. Guo Q, Zheng Y, Shi J, Wang J, Li G, Li C, et al. Immediate psychological distress in quarantined patients with COVID-19 and its association with peripheral inflammation: a mixed-method study. Brain Behav Immun. 2020;88:17–27. https://doi.org/10.1016/j.bbi.2020.05.038.
228. Najjar S, Najjar A, Chong DJ, Pramanik BK, Kirsch C, Kuzniecky RI, et al. Central nervous system complications associated with SARS-CoV-2 infection: integrative concepts of pathophysiology and case reports. J Neuroinflammation. 2020;17(1):231. https://doi.org/10.1186/s12974-020-01896-0.
229. Troyer EA, Kohn JN, Hong S. Are we facing a crashing wave of neuropsychiatric sequelae of COVID-19? Neuropsychiatric symptoms and potential immunologic mechanisms. Brain Behav Immun. 2020;87:34–9. https://doi.org/10.1016/j.bbi.2020.04.027.
230. Lorkiewicz P, Waszkiewicz N. Biomarkers of post-COVID depression. J Clin Med. 2021;10(18):4142. https://doi.org/10.3390/jcm10184142.
231. Bhargava A, Fukushima EA, Levine M, Zhao W, Tanveer F, Szpunar SM, et al. Predictors for severe COVID-19 infection. Clin Infect Dis. 2020;71(8):1962–8. https://doi.org/10.1093/cid/ciaa674.
232. Lagunas-Rangel FA. Neutrophil-to-lymphocyte ratio and lymphocyte-to-C-reactive protein ratio in patients with severe coronavirus disease 2019 (COVID-19): a meta-analysis. J Med Virol. 2020;92(10):1733–4. https://doi.org/10.1002/jmv.25819.
233. Liu Q, Dai Y, Feng M, Wang X, Liang W, Yang F. Associations between serum amyloid a, interleukin-6, and COVID-19: a cross-sectional study. J Clin Lab Anal. 2020;34(10):e23527. https://doi.org/10.1002/jcla.23527.
234. Yuan B, Li W, Liu H, Cai X, Song S, Zhao J, et al. Correlation between immune response and self-reported depression during convalescence from COVID-19. Brain Behav Immun. 2020;88:39–43. https://doi.org/10.1016/j.bbi.2020.05.062.
235. Mazza MG, Palladini M, De Lorenzo R, Magnaghi C, Poletti S, Furlan R, et al. Persistent psychopathology and neurocognitive impairment in COVID-19 survivors: effect of inflammatory biomarkers at three-month follow-up. Brain Behav Immun. 2021;94:138–47. https://doi.org/10.1016/j.bbi.2021.02.021.
236. Forget M-F, Del Degan S, Leblanc J, Tannous R, Desjardins M, Durand M, et al. Delirium and inflammation in older adults hospitalized for COVID-19: a cohort study. Clin Interv Aging. 2021;16:1223–30. https://doi.org/10.2147/CIA.S315405.
237. Zheng F, Xie W. High-sensitivity C-reactive protein and cognitive decline: the English longitudinal study of ageing. Psychol Med. 2018;48(8):1381–9. https://doi.org/10.1017/S0033291717003130.
238. Vintimilla R, Hall J, Johnson L, O'Bryant S. The relationship of CRP and cognition in cognitively normal older Mexican Americans: a cross-sectional study of the HABLE cohort. Medicine (Baltimore). 2019;98(19):e15605. https://doi.org/10.1097/MD.0000000000015605.
239. Mitko A, Rothlein D, Poole V, Robinson M, McGlinchey R, DeGutis J, et al. Individual differences in sustained attention are associated with cortical thickness. Hum Brain Mapp. 2019;40(11):3243–53. https://doi.org/10.1002/hbm.24594.

# Part II

# Inflammation and Specific Disorders

# Stress and Kynurenine-Inflammation Pathway in Major Depressive Disorder

8

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**Abstract**

Major depressive disorder (MDD) is one of the most prevalent disorders and causes severe damage to people's quality of life. Lifelong stress is one of the major villains in triggering MDD. Studies have shown that both stress and MDD, especially the more severe conditions of the disorder, are associated with inflammation and neuroinflammation and the relationship to an imbalance in tryptophan metabolism towards the kynurenine pathway (KP) through the enzymes indoleamine-2,3-dioxygenase (IDO), which is mainly stimulated by pro-inflammatory cytokines and tryptophan-2,3-dioxygenase (TDO) which is activated primarily by glucocorticoids. Considering that several pathophysiological mechanisms of MDD underlie or interact with biological processes from KP metabolites, this chapter addresses and discusses the function of these mechanisms. Activities triggered by stress and the hypothalamic-pituitary-adrenal (HPA) axis and immune and inflammatory processes, in addition to epigenetic phenomena and the gut-brain axis (GBA), are addressed. Finally, studies on the function and mechanisms of physical exercise in the KP metabolism and MDD are pointed out and discussed.

**Keywords**

Stress · Glucocorticoids · Kynurenine pathway · Neuroinflammation · Autonomic nervous system · Major depressive disorder

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## 8.1 Introduction

Major depressive disorder (MDD) affects 300 million people worldwide. It contributes to suicides and morbidity, causing a significant loss of quality of life [1–3]. Depressive people are at high risk of committing suicide, one of the leading causes of death worldwide [1]. The disturbance of serotonin (5-HT) metabolism in the brain is linked to MDD, suicide, and alcohol use disorder [4].

Monoaminergic and glutamatergic neurotransmission and the immune system are biological systems involved in the pathophysiology of MDD. A range of studies has shown that the kynurenine pathway (KP) is an intersection point in the relationship between these three systems. Neuroactive KP metabolites are involved in the interface between immune function and serotonergic neurotransmission, culminating in the positive or negative modulation of glutamatergic neurotransmission and, thus, inducing neuroprotection or neurotoxicity. The literature provides evidence that an imbalance in KP metabolites is involved in several neuropsychiatric disorders, including MDD [5].

Tryptophan (TRP) is an essential amino acid. That means it is not synthesized in the human body. This amino acid is present in foods such as eggs, milk, meat, soybean, potatoes, and cereals. After absorption, it is carried with albumin (80–90%) and free form in the blood. TRP is related to appetite, sleep-wake rhythm, and pain perception [6].

Kynurenine (KYN) is a metabolization product of L-tryptophan (L-Tr). Similarly, 5-HT is another product of L-Tr metabolization, which is initially converted to 5-hydroxytryptophan and then to 5-HT. The proportion of 5-HT and KYN after these two different pathways is, respectively, 5% and 95%. The imbalance of this proportion is related to the pathogenesis of depressive-related disorders mediated by 5-HT deficiency. Both substances are produced centrally and peripherally [7, 8].

The KP is the primary degradation pathway of TRP and plays a role in the immune response as it comprises compounds that play a role in the nervous system. These substances have neuroprotective or neurotoxic effects. Under physiological conditions, substances related to the KP present a balance between neuroprotective and neurotoxic activities, but in cases of stress and exacerbated immune activation, there is an imbalance that increases the excitotoxic activity of metabolites in the N-methyl-D-aspartate (NMDA) receptor [9].

Studies emphasize that the link between inflammation markers and activation of the KP is enhanced in cases where the immune system has exacerbated activation. With an overactive immune system, a high rate of pro-inflammatory cytokines is released, which have as one of their functions to stimulate the conversion of TRP to KYN in the periphery. The KYN crosses the blood-brain barrier (BBB), increasing pro-inflammatory substances and KYN metabolites in the central nervous system (CNS). Thus, creating a vicious and harmful cycle for body homeostasis can culminate in various illnesses and psychiatric disorders, as MDD [10].

Neuronal plasticity is a neuronal adaptation mechanism, and its function is affected by MDD. The exacerbated immune activation present in the disorder's

pathophysiology causes alterations in the function of the hippocampus, prefrontal cortex (PFC), and amygdala and in the glutamate and glucocorticoid pathways [11].

Several mechanisms related to the KP are inherent to the pathophysiology of MDD, such as the stimulation of the immune system and the effects on neuronal plasticity. The changes in brain plasticity seem to be one of the main morphophysiological processes in depression. Therefore, this study considers the mechanisms that interact or are underlying stress, KP metabolism, and its relationship with MDD.

## 8.2 Kynurenine Pathway

Among the theories that try to explain the pathophysiology of MDD are monoaminergic and inflammatory. These two theories are not watertight. In the KP, monoaminergic and inflammatory mechanisms are related by some intersections [12]. In the KP, indoleamine-2,3-dioxygenase (IDO), which can be found in extrahepatic tissues, including the brain, together with tryptophan-2,3-dioxygenase (TDO), is responsible for degrading the TRP [13]. IDO can be stimulated by pro-inflammatory cytokines, lipopolysaccharides (LPSs), and free radicals [14, 15] whereas TDO is mainly activated by glucocorticoids [14] and is restricted to liver tissue [16]. Peripheral cell-mediated immune activation and inflammation may cause microglial activation with increased levels of pro-inflammatory cytokines [14].

The activity of the IDO or TDO enzymes promotes the transformation of TRP into N-formylkynurenine (NFK) [13], which is metabolized by formamidase, the second enzyme in the KP to produce KYN [17, 18]. KYN metabolism occurs mainly in astrocytes and microglia, each of which leads KYN to follow a different pathway, considering that the kynurenine monooxygenase (KMO) is expressed in microglia but not in astrocytes [19], and kynurenine aminotransferases (KATs) enzymes are expressed in astrocytes but not in microglia [20].

Through the action of KATs, KYN undergoes irreversible transamination to form kynurenic acid (KYNA) [20]. Four KATs appear to catalyze the KYN reaction to KYNA. However, KAT II is believed to be the major biosynthetic enzyme [21, 22]. KYNA is a glutamatergic NMDA receptor antagonist [17, 23, 24] and is therefore considered a neuroprotective metabolite [25, 26]. However, the activity of KATs does not appear to be able to compete with the other arm of the KP, the direct pathway for the production of quinolinic acid (QA), by transforming KYN into its metabolites [18].

KYNA is a competitive broad-spectrum glutamate receptor antagonist, inhibiting all three ionotropic excitatory amino acid receptors (NMDA, kainate, and AMPA) [27]. This metabolite also has a greater affinity for the glycine obligate co-agonist site of the NMDA receptor [28]. Besides, KYNA is a noncompetitive α7 nicotinic acetylcholine (ACh) receptor inhibitor [29]. Given this, increased concentrations of KYNA decrease extracellular levels of glutamate and dopamine in various regions of the rat brain [30–32]. In addition, KYNA regulates the ACh levels in the medial PFC of male rats, thereby showing the ability to attenuate extracellular ACh levels [33].

By another KP, KMO and kynureninase (KYNU) catalyze the degradation of KYN to 3-hydroxykynurenine (3-HK) and anthranilic acid (AA), respectively [34, 35]. 3-HK can stimulate the production of reactive oxygen species (ROS) and cellular apoptosis [36, 37]. In the CNS, 3-HK conversion occurs mainly in microglia, where it will later be transformed into 3-hydroxyanthranilic acid (3-HAA) by the action of KYNU, considering that KYNU preferentially recognizes 3-HK concerning KYN, thus catalyzing the formation of 3-HAA [18, 38]. 3-HAA generates highly reactive hydrogen peroxide and hydroxyl radicals [39]. After the production of 3-HAA, there are two possible degradation pathways. One pathway proceeds with the complete oxidation of 3-HAA to form adenosine triphosphate (ATP) and a small amount of picolinic acid (PA) through the action of 2-amino-3-carboxymuconate-6-semialdehyde decarboxylase (ACMSD). The other pathway promotes the oxidation of 3-HAA by 3-hydroxyanthranilic acid oxidase (3-HAO) into 2-amino-3-carboxymuconic semialdehyde (ACMSA). ACMSA spontaneously is converted to QA [40, 41]. QA is responsible for selectively activating NMDA receptors, and thus, persistent activation of excitatory neurons causes excitotoxicity [42]. QA is processed by quinolinic acid phosphoribosyltransferase (QPRT) to the nicotinamide adenine dinucleotide + (NAD+) precursor nicotinic acid mononucleotide (NAMN) [43–45]. Then, through a reaction catalyzed by NAMN adenylyltransferases (NMNATs), the NAMN is converted to nicotinic acid adenine dinucleotide (NAAD). Finally, through the action of glutamine-dependent NAD+ synthetase (NADsyn), the NAAD metabolite is converted to NAD+ [40]. KP is summarized in Fig. 8.1.

Thus, KP inhibition can be therapeutic in neuroinflammatory situations by reducing the production of excitotoxins such as QA. From KP, 3-HK is responsible for generating free radicals and causing neuronal apoptosis. In its turn, 3-HAA generates highly reactive hydrogen peroxide and hydroxyl radicals. The QA selectively activates NMDA receptors, promoting high concentrations of extracellular glutamate and persistent activation of excitatory neurons, causing excitotoxicity. Given this, the accumulation of QA can result in neuronal excitotoxicity and selective apoptosis of astrocytes. Thus, it can subsequently promote neurodegenerative changes, making them susceptible to MDD [16, 46, 47].

Glutamate reuptake occurs through excitatory amino acid transporter 2 (EAAT2), the leading glutamate transporter in the brain, predominantly expressed in astrocytes [48]. In astrocyte cell cultures, QA decreased glutamate uptake, contributing to increased extracellular concentrations, collaborating with the super stimulation of the glutamatergic system [45]. From activation of NMDAR, Na+ and Ca2+ influx occur, increasing Ca2+ cytoplasmic levels and uptake in the mitochondria. Elevated mitochondrial Ca2+ levels can induce ROS rise and inhibit ATP generation (Fig. 8.2) [49].

On the other hand, KP metabolism can also be cytoprotective through intracellular NAD+ synthesis [50]. In genomic DNA, poly-(ADP-ribose) polymerase (PARP) are DNA-binding enzymes, particularly PARP-1, activated by free radical-mediated DNA strand breaks and play a crucial role in excision repair. NAD+ is the sole substrate for the PARP [51, 52].

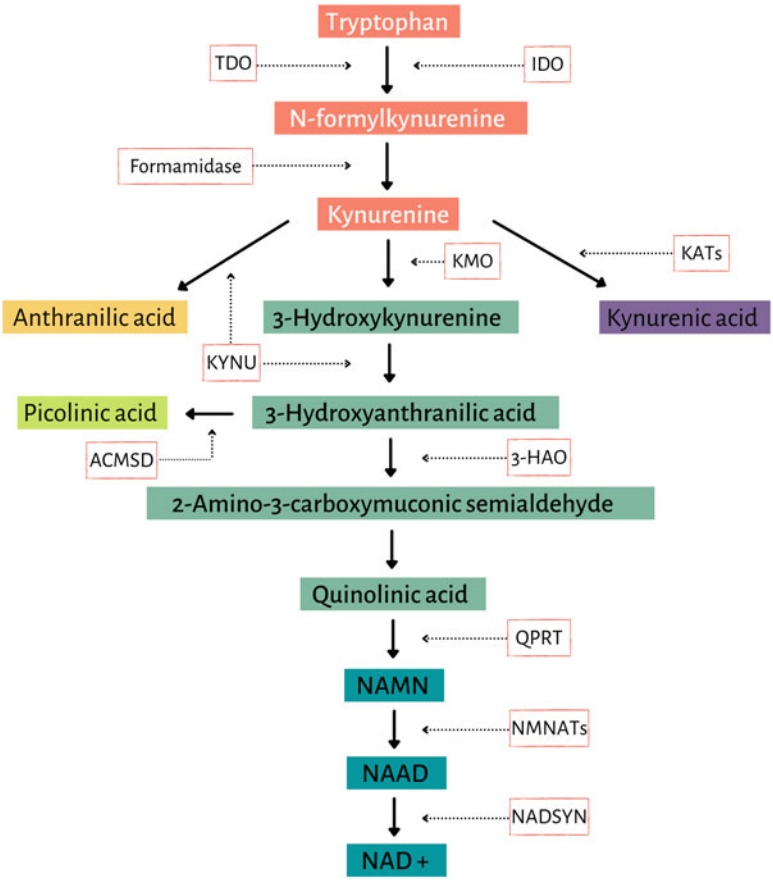


**Fig. 8.1** Kynurenine pathway: tryptophan (TRP) is degraded either by indoleamine-2,3-dioxygenase (IDO), in extrahepatic tissues, or by tryptophan-2,3-dioxygenase (TDO), in the liver. These two enzymes convert TRP into N-formylkynurenine (NFK), which is metabolized by formamidase to form kynurenine (KYN). In astrocytes, kynurenine aminotransferases (KATs) catalyze KYN to kynurenic acid (KYNA), the neuroprotective metabolite. In this pathway, anthranilic acid (AA) is also formed by kynureninase (KYNU). In addition, in microglia, kynurenine 3-monooxygenase (KMO) converts KYN in 3-hydroxykynurenine (3-HK). Subsequently, 3-HK is transformed into 3-hydroxyanthranilic acid (3-HAA) by KYNU. 3-HAA can be oxidized to either picolinic acid (PA) or 2-amino-3-carboxymuconic semialdehyde (ACMSA) by 2-amino-3-carboxymuconate-6-semialdehyde decarboxylase (ACMSD) and 3-hydroxyanthranilic acid oxidase (3-HAO), respectively. Then ACMSA spontaneously converts to quinolinic acid (QA). QA is processed by QA phosphoribosyltransferase (QPRT) to the NAD+ precursor nicotinic acid mononucleotide (NAMN), which is then converted to nicotinic acid adenine dinucleotide (NAAD) via NAMN adenylyl transferases (NNMATs). Finally, through the action of glutamine-dependent NAD+ synthetase (NADsyn), the NAAD metabolite is converted to NAD+. Images were extracted from the BioRender app

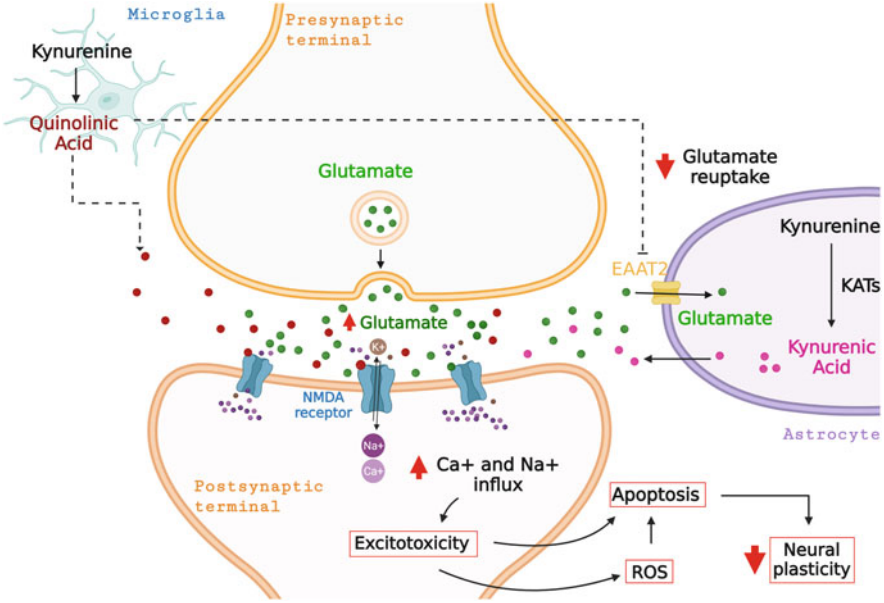


**Fig. 8.2** Quinolinic acid and glutamate after microglial activation induced by pro-inflammatory cytokines, increased quinolinic acid (QA) levels can be found in the extracellular domain. The release of kynurenic acid (KYN), the glutamatergic N-methyl-d-aspartate (NMDA), receptor antagonist, happens in astrocytes. QA activates the NMDA receptor. QA also inhibits the glutamate reuptake by astrocytes carried out through excitatory amino acid transporter 2 (EAAT2), increasing extracellular glutamate. QA and glutamate are then free to stimulate the NMDA receptor repeatedly. Thus, there is an increase in calcium ($Ca^{2+}$) and sodium ($Na^{+}$) influx. Elevated cytoplasmic levels of $Ca^{2+}$ can induce mitochondrial $Ca^{2+}$ uptake, stimulating the production of reactive oxygen species (ROS) while inhibiting the production of adenosine triphosphate (ATP) and leading to cells apoptosis and damaged neural plasticity. Images were extracted from the BioRender app

## 8.3 Kynurenine Pathway and Inflammation

The currently most accepted hypothesis supports the claim that metabolites of the KP are related to inflammation associated with depression through their effect on glutamate receptors in the brain [53]. Another mechanism related to KP and MDD involves IDO, which mediates the production of pro-inflammatory substances for the breakdown of TRP in KYN both in the periphery and in the CNS [23]. The substances produced by KP have different functions in the immune system. AA and KYNA have anti-inflammatory functions, and QA is a pro-inflammatory substance that stimulates mitochondrial dysfunction, cell death, apoptosis, energetic deficit, and lipid peroxidation, among other neurotoxic mechanisms [42].

LPS induces the expression of pro-inflammatory cytokines that activate IDO1, causing a sustained immune activation, culminating in depression, anxiety, or

cognitive deficits. The chronic inhibition of IDO1 or TDO2 seems to reverse the effects induced by LPS [54].

The CSF of patients with an acute neuroinflammatory disease had higher KYN, QA, and AA levels and lowered TRP, 3HAA, and KYNA. Furthermore, the nitric oxide (NO) pathway and neopterin were also altered in patients compared to controls. In the NO pathway, the levels of arginine and citrulline were decreased, and asymmetric dimethylarginine and argininosuccinic acid were increased. The neopterin was significantly elevated [55].

NO and neopterin are substances present at high levels in neurological diseases with inflammatory mechanisms. There is a correlation between elevated neopterin and the KYN/TRP ratio in serum and CSF [56, 57]. The levels of neopterin and interferon (IFN)-γ inducible protein of 10 kDa (IP-10) stimulates IFN-γ activity, which is shown to activate the KP in patients with acute CNS infection, mainly mediating neurotoxic effects [58]. Individuals with advanced Parkinson's have increased neopterin formation and increased TRP degradation, corroborating the hypothesis of a relationship between the two pathways [56].

IDO mediates the depletion of TRP in the KP through IFN-γ. An in vitro study on the human epithelial cell line RT4 found that INF stimulation inhibited the growth of *Staphylococcus aureus* mediated by IDO, an effect abolished by endogenous NO. Both endogenous and exogenous NO reduce the IDO level in RT4 cells. This effect does not occur due to a decrease in IDO gene transcription or mRNA stability but because NO production leads to degradation accelerated IDO in the proteasome [58].

In an experimental human protocol, healthy individuals submitted to an immune challenge presented depressive symptoms and increased KYN, TRP, KYNA, IL-6, and TNF-α in parallel to a higher plasma IDO function measured by KYN/TRP ratio [59].

Bipolar disorder (BD) is a condition involving the immune system's chronic activation. Some individuals do not respond to the traditional treatments available and are then diagnosed with treatment-resistant bipolar depression (TRBDD). Individuals TRBDD were treated with a combination of escitalopram and the cyclooxygenase-2 (COX-2) inhibitor celecoxib (CBX) as an add-on. The research results suggest high levels of IL-1β in TRBDD individuals, culminating in an increase in pro-inflammatory cytokines through the COX-2 pathway with activation of KP [60].

Chronic low-grade inflammation is present in individuals with primary hyperparathyroidism. A cross-sectional survey found that individuals with primary hyperparathyroidism without other acute diseases have KP activation. Circulating concentrations of KYN and QA were related to C-reactive protein. A critical relationship occurred between TRP, KYN, and QA with echocardiographic parameters of cardiac remodeling, an important alteration present in individuals with primary hyperparathyroidism. High QA levels were associated with the left ventricular mass index, left ventricular hypertrophy, and left atrial volume index [61].

It is possible to verify that several inflammatory diseases are related to alterations in KP and their respective metabolites. It is considered a key mechanism for developing and progressing the diseases mentioned above and MDD.

## 8.4 Stress and Hypothalamic-Pituitary-Adrenal (HPA) Axis and Kynurenine Pathway

Psychosocial stress is one of the risk factors for mood disorders. Stressful aversive stimuli are involved in the complex mechanisms of MDD pathogenesis [62, 63]. Acute stress increases blood levels of chemokines and cytokine [64] whereas chronic stress promotes the sensitization of inflammatory responses to stress [65]. Blood levels of pro-inflammatory monocytes and cytokines are increased from psychosocial stress. In addition, stress also increases monocyte circulation to the brain and microglial activation in the amygdala, hippocampus, and prefrontal cortex [15, 66, 67].

Stress can affect the activity of enzymes that regulate the KP [68]. Among the enzymes is IDO, which is upregulated by pro-inflammatory cytokines such as TNF-α, IFN-γ, and IL-6 [14, 68, 69]. The association of stress with inflammation, increasing IDO activity, culminates in increased KYN levels and reduced availability of TRP for the 5-HT pathway [65].

Some studies provide evidence on the relationship of KP hyperactivity with stress-related disorders such as MDD [14, 70–75]. Rats subjected to acute stress showed alterations in the gene expression of enzymes that regulate the KP in a regionally dependent manner. There was an increase in IDO, KMO, and KYNU mRNA expression in the amygdala after exposure to acute stress. A reduction in KYNU mRNA gene expression was observed in the medial prefrontal cortex. An increase in TDO mRNA expression was observed in the hypothalamus of rats after exposure to acute stress. On the other hand, there was a reduction in the expression of TDO mRNA in the hippocampus. The identification and understanding of these regional differences regarding the KP can provide further elucidation about the dysregulated mechanisms in MDD and other stress-related disorders [76].

Chronic moderate stress (CMS) in rats culminated in depressive-like behavior. Additionally, stressed animals showed an increase in TNF-α and IL-1β levels and IDO expression in the frontal cortex. CMS animals also showed increased QA levels, indicating an increase in IDO activity [12]. Chronic psychosocial stress in mice, through a protocol of chronic social defeat (CSD), induced higher blood levels of TNF-α, IFN-γ, KYN, 3-HK, and KYNA. KYN and 3-HK levels also increased in the amygdala and hippocampus of stressed animals. Inhibition of IDO reversed CSD-induced levels of KYNs [65].

Stress-induced activation of the HPA axis [76] results in the release of glucocorticoids from the adrenals which can lead to the induction of TDO upon activation of intracellular glucocorticoid receptors (GR) [77].

There is an increase of TDO activity after dexamethasone administration in hepatocytes [78] and a lowering of TRP availability [79]. A meta-analysis knew

that depressive individuals had reduced levels of TRP compared with nondepressed individuals [80]. In a study with dates of depressed individuals involving the analyses of KYN levels, TRP, and cortisol, no differences between KYN, TRP, or KYN/TRP ratio in depressive versus nondepressive individuals were found. However, there was found enhancement of evening cortisol in individuals with decreased KYN/TRP ratio in the total sample. The hypothesis for this phenomenon is that the enhanced levels of cortisol cause glucocorticoid resistance, downregulating glucocorticoid receptor and then reducing the signalization to increase TDO by cortisol [81, 82]. However, this is just theoretical analysis, and then more studies are necessary to support this hypothesis.

For individuals with high cognitive reactivity marked as dysfunctional cognition after stressful circumstances or sad mood, the supplementation with TRP-rich hydrolyzed protein reduces the negative mood response to stress and the levels of cortisol after stress exposure [83].

In suicidal MDD patients, lower TRP levels and increased KYN/TRP rates were observed compared to healthy controls and non-suicidal MDD. MDD without suicide risk also presented an increased KYN/TRP rate than controls. Plasma levels of KYN were higher in patients with a history of suicide attempts. Plasma cortisol levels were significantly higher in MDD patients with and without suicide risk and were positively correlated with KYN levels and the KYN/TRP ratio [84]. In suicidal MDD patients, cytokine activation markers were positively correlated with the KYN/TRP rate [75].

Still considering the relationship of the HPA axis with the KP, it is interesting to observe the studies in which healthy male volunteers were treated acutely with gamma-hydroxybutyrate (GHB), a GABA metabolite and $GABA_B$ receptor agonist, which is used in narcolepsy. In the morning, after nocturnal administration, subjects treated with GHB had reduced KP metabolites such as KYN, KYNA, 3-HK, QA, as well as reduced 3-HK/KYNA rates and cortisol-awakening response. Thus, GHB is suggested as a potential stress reducer and antidepressant [85]. It was hypothesized that GHB decreases cortisol levels by anti-unknown depressant mechanisms and lowers inflammatory cytokines like IFN-γ and TNF-α, reducing TDO and IDO, respectively [78, 85, 86]. The relationship of stress with the HPA axis and KP and inflammation and depression is illustrated in Figs. 8.3, and 8.4.

## 8.5 Kynurenine Pathway and Major Depressive Disorder

Protocols for studies in humans with MDD have observed imbalances in the KP, often with an increase in neurotoxic metabolites, associated with an increase in inflammatory markers. MDD inpatients had reduced levels of KYNA and mean TRP index, in addition to an increase in TRP degradation and a reduction in neuroprotective rate, measured by plasma KYNA levels over KYN levels. In the same series of studies, the authors found that the neuroprotective rate increased after 6 weeks of antidepressant treatment, but only in patients admitted after the first depressive episode [47]. Still considering the imbalance of the KP pathway, a recent

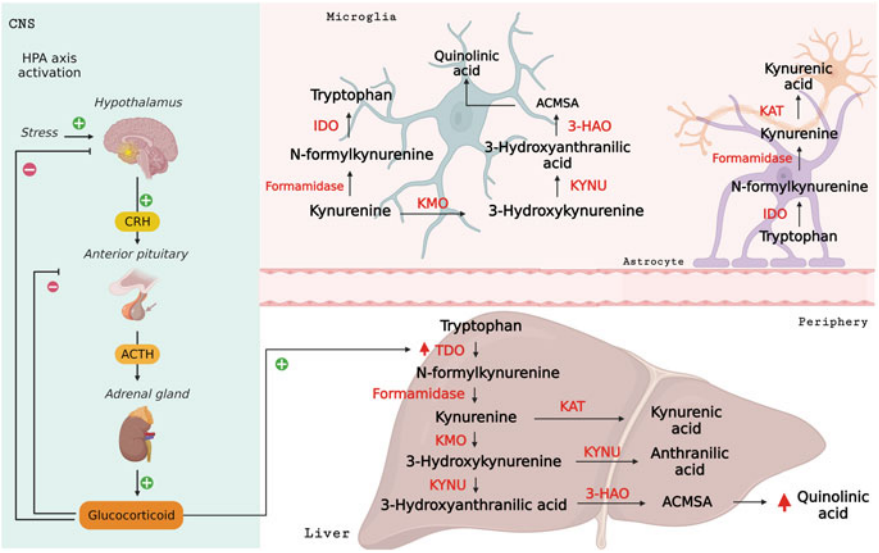


**Fig. 8.3** Stress and kynurenine pathway: the hypothalamic-pituitary-adrenal (HPA) axis is increased by stress, raising the release of corticotrophin release hormone (CRH), which in turn activates the secretion of adrenocorticotrophin hormone (ACTH) by the pituitary. ACTH then stimulates the secretion of glucocorticoids from the adrenal cortex. The glucocorticoid increases the tryptophan-2,3-dioxygenase (TDO) activation. TDO metabolizes tryptophan to kynurenine, which is then converted to kynurenic acid (KYNA) by kynurenine aminotransferase (KAT) or 3-hydroxykynurenine by kynurenine monooxygenase (KMO). 3-hydroxykynurenine (3-HK) is further metabolized to anthranilic acid (AA) or 3-hydroxyanthranilic acid (3-HAA) by kynureninase (KYNU). 3-HAA can be oxidized to 2-amino-3-carboxymuconic semialdehyde (ACMSA) by the action of 3-hydroxyanthranilic acid oxidase (3-HAO). Then ACMSA spontaneously is converted to quinolinic acid (QA). Images were extracted from the BioRender app

meta-analysis highlights the shift in the metabolization of the pathway, showing that there is an increase in KYN over TRP and an increase in neurotoxic, to the detriment of neuroprotective metabolites [87].

The KP appears to be involved with the suicide risk or attempted in depressed patients. Adolescents who attempted suicide showed reduced levels of TRP and higher levels of KYN than TRP [88]. These results indicate a strong relationship between the KP and suicidal behavior in patients with MDD. The enzyme ACMSD appears to protect against suicidal behavior through a balance of KYN metabolites. ACMSD activity reduces the formation of QA through a competitive synthesis of the neuroprotective metabolite PA. One study observed that individuals with suicidal behavior had lower PA levels and a lower PA/QA rate, both in peripheral blood and CSF. Individuals with the genotype for lower ACMSD expression had a higher prevalence of suicide attempts associated with higher levels of QA in the CSF [89].

Electroconvulsive therapy (ECT) is considered the standard gold treatment for acute and treatment-resistant depression (TRD), as it controls the increased functional connectivity of specific neural networks present in disorder [90]. ECT

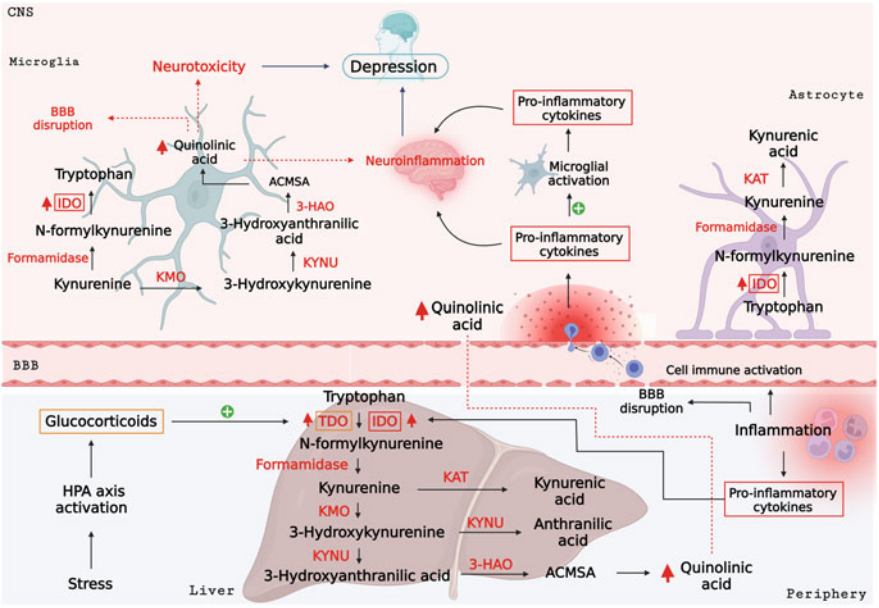


**Fig. 8.4** Kynurenine pathway and neuroinflammation and depression: tryptophan-2,3-dioxygenase (TDO) is mainly activated by glucocorticoids, whose release increases in situations of stress and depression. Peripheral cell-mediated immune activation promotes pro-inflammatory cytokines release. These cytokines stimulate indoleamine-2,3-dioxygenase (IDO) and can also stimulate microglial activation, increasing pro-inflammatory cytokines levels in the central nervous system (CNS). IDO and TDO exacerbated stimulation shifts metabolism to the kynurenine pathway (KP) at the expense of the serotonin pathway. The increase in kynurenine (KYN) levels in these situations can increase quinolinic acid (QA) and decrease kynurenic acid (KYNA). QA activates N-methyl-D-aspartate (NMDA) receptor and increases extracellular glutamate, leading to neurotoxicity. Peripheral QA cannot cross the Blood-Brain Barrier (BBB) under physiological conditions but inflammation can disrupt the BBB, opening the way for the peripheral QA into the CNS. The CNS QA accumulation can result in neuronal excitotoxicity as it is also capable of stimulating mitochondrial dysfunction, cell death, apoptosis, energetic deficit, lipid peroxidation, among other toxic mechanisms. Thus, the imbalance in tryptophan metabolism contributes to the triggering and severity of depression. Images were extracted from the BioRender app

decreased plasma levels of QA, KYN, and TRP in TRD patients. Besides, the QA/KYNA ratio significantly decreased in TRD patients from ECT [91]. This evidence suggests that the balance of KP metabolites is a mechanism involved in the ECT therapeutic effect.

Women diagnosed with peripartum depression had increased plasma levels of IL-6 and IL-8 and reduced 5-HT, IL-2, and QA. These changes were also associated with the severity of symptoms and increased risk of suicide, indicating that the pathophysiology of peripartum depression involves dysregulation of the immune system and, inherently, of the KP [92].

Some studies provide evidence that alterations in the KP are underlying the brain gray matter volume and neural circuits changes in MDD individuals [93]. A study

with magnetic resonance imaging (MRI) scan observed that MDD subjects had a reduction in the thickness of areas of the medial prefrontal cortex (mPFC), concomitantly with a reduction in the serum concentrations rate of KYNA/3HK and log KYNA/QA, suggesting that an imbalance between neuroprotective and neurotoxic metabolites of the KP are related to the loss of brain gray matter [94]. In another study, researchers observed lower hippocampal and amygdala volume and lower serum KYNA/3HK and KYNA/QA rates in unmedicated MDD subjects. The KYNA/QA rate was negatively correlated with anhedonia and positively correlated with hippocampal and amygdala volumes, suggesting dendritic atrophy and anhedonia associated with depression, from an imbalance between neuroprotective and neurotoxic metabolites of KYN [73].

In addition to contributing to structural and functional impairments and mood changes, the KP metabolism is involved with cognitive deficits and sleep disturbance associated with MDD. Women with MDD had a negative association between verbal learning and KYN levels. Furthermore, processing speed and verbal and visual learning were negatively correlated with the KYN/TRP rate [95]. Sleep disturbances in MDD subjects were associated with reduced KYNA/QA and increased C-reactive protein levels in the serum of patients [96].

Considering that neurotoxic metabolites of the KP are related to suicide risk, it is also essential to observe the studies, which suggest that these metabolites and the imbalances towards possible neurotoxicity are associated with higher inflammatory markers present more frequently in melancholic symptoms that appear in severe depression with psychotic and suicidal features. In this sense, the studies by Milaneschi et al. [97] observed that plasma KYN and QA levels, as well as KYN/TRP, QA/KYN, and QA/KYNA rates, were positively associated with TNF and CRP, which were higher in individuals with melancholic symptoms and atypical symptoms related to energy. These results align with results from other studies, which showed a reduction in KYNA and the KYNA/QA ratio in depression with psychotic characteristics [98].

In patients diagnosed with MDD and not medicated, KYN was increased in plasma and cerebrospinal fluid (CSF) associated with increased levels of plasma tumor necrosis factor (TNF). A higher level of KYN concerning TRP is associated with high levels of KYN, QA, and KYNA in the CSF. In a subgroup of individuals, increased levels of TNF and KYN, compared to plasma TRP levels, was associated with greater severity of depression, anhedonia, and nonresponse to treatment [53].

In a series of studies looking at individuals at high risk for MDD and mice undergoing a chronic stress protocol, the authors observed increased serum AA levels and decreased TRP concentrations. Based on the evidence in these studies, the researchers suggest AA as a sensitive biomarker that can be used to detect MDD risk in individuals from susceptible groups [99].

Studies with animal protocols also show peripheral and brain alterations in the metabolites of the KP, associated with depressive-like behaviors. In a protocol with an immune challenge through the administration of LPS, the dose that induced depressive-like behavior in mice also reduced neuroplasticity in the hippocampus and increased the neurotoxic metabolite 3-HK in the hippocampus and cerebral

cortex [5]. Mild chronic stress in mice induced depressive-like behavior and, in parallel, increased KP metabolism, culminating in high levels of 3HK in peripheral blood. In the same studies, a reduction in TRP was observed in the hippocampus and striatum of animals subjected to stress [100].

It is worth highlighting the studies that suggest KMO knockout mice as a model that can meet the three validity criteria for a suitable MDD animal model. In these studies, KMO knockouts showed increases in KYN, KYNA, and AA in serum and hippocampus, in addition to a reduction in hippocampal 5-HT turnover. At the same time, the animals showed depressive-like behavior, which was reversed after chronic administration of imipramine and sertraline [101].

Also noteworthy are recent studies by Tanaka et al. [102] who observed a potent antidepressant-like effect of KYNA intracerebroventricularly in mice. Worthy of mention, KYNA seems to have interacted strongly with the 5-HT2 serotonergic receptor; weakly with the D2, D3, and D4 dopaminergic receptors; and moderately with the GABAergic, γ-aminobutyric acid ($GABA_A$) receptor to exert its antidepressant-like effect.

## 8.6 Kynurenine Pathway, Autonomic Nervous System, and MDD

Stress activates the autonomic nervous system (ANS) [16, 103–105]. Dysregulation of the ANS is one of the potential reasons for the relationship between depression and heart problems [106–109]. In chronic stress situations, such as MDD, the sympathetic nervous system (SNS) can be continuously activated without the counteracting action of the parasympathetic nervous system (PNS). Thus, the immune system can be activated, culminating in increased levels of pro-inflammatory cytokines [16, 105].

Under stress, the paraventricular nucleus of the hypothalamus (PVN) increases the release of corticotropin-releasing hormone (CRH). CRH-containing neurons project to noradrenergic centers in the brainstem and spinal cord. Noradrenergic nuclei send projections to preganglionic neurons, increasing sympathetic and decreasing parasympathetic activities. Persistent sympathetic nervous system activation stimulates CRH release, culminating in an endless loop [16, 110–113].

The SNS is persistently activated in prolonged stress and MDD without SNP antagonism. Epinephrine and norepinephrine, through α and β adrenoceptors, increase the release of pro-inflammatory cytokines, such as TNF, IL-1, IL-6, and IFNs. Pro-inflammatory cytokines induce IDO activity, leading to increased KYN production from TRP. Following the KP, there is an increase in the production of metabolites, such as 3-HK, 3-HAA, and QA [16, 113, 114].

The reduction in PNS activity culminates in an alteration in the immune response through the cholinergic anti-inflammatory pathway. Autonomic imbalance with reduced cholinergic activity is implicated in psychiatric disorders in comorbidities with cardiovascular disease and significant variations in the immune system. Among the inflammatory variations is the increased release of TNF-α. Outstanding that the

mechanisms are not straightforward and interact with each other. Thus, an imbalance of the KP from hyper inflammation or an imbalance of the ANS can increase KYNA release. Although KYNA is a metabolite that provides more protection due to its glutamatergic antagonist and anti-inflammatory action, its nicotinic cholinergic antagonist action contributes to an increase in inflammation through the imbalance of autonomic function [115].

## 8.7 Epigenetic, Kynurenine Pathway, and MDD

Mice exposed to Bacille Calmette-Guérin (BCG) exhibited increased depressive-like behavior, but rats knocked out to the IDO gene or pre-treated with an IDO inhibitor completely had blocked BCG-induced depressive-like behaviors. The IDO and 3-HAO mRNA expressions were increased, as well as TNF-α and IFN-γ, after exposure to BCG. These findings suggest that an epigenetic increment of IDO and 3-HAO expression is probably mediated by TNF-α and IFN-γ [116]. Another study observed that mice submitted to an immunological challenge with BCG exhibited depressive-like behavior, unlike IDO1 knockout mice who submitted to the same challenge. At the same time, wild mice had a higher number of replication-dependent acetylated histones in microglia after recovery from inflammation than IDO1 deficient animals. In addition to differences in posttranslational epigenetic changes, wild-type and IDO1 knockout mice subjected to immune challenge showed differences in the expression of several proteins, suggesting that inflammation and microglial activation promote IDO-dependent molecular changes, which may be responsible for the duration of depressive behavior [117].

The induction of IDO was observed in mouse microglia cell culture in the presence of IFN-γ. When IL-4 was included in the culture containing IFN-γ, an average of 3.2-fold in KYN concentration was observed compared to IFN-γ alone. A synergistic action was found between IFN-γ, IL-4, and IL-13 in IDO induction. First, neither increase in IDO was found in cultures with IL-4 or IL-13. Although, in microglia culture with IFN-γ, an increase about sixfold was found in IDO mRNA expression. Finally, the association IFN-γ plus IL-4 resulted in an increase of 2-fold and IL-13, 3.2-fold compared with IFN-γ alone, showing synergic relations between IFN-γ and anti-inflammatory IL-4 and IL-13 cytokines [118].

The tryptophanyl tRNA synthetase (TrpRS) is an enzyme that attaches TRP to tRNA to generate proteins. The expression of this enzyme enhances in mouse microglia with INF-γ stimulation but declines with IFN-γ plus IL-4 or IL-13. IL-4 or IL-13 alone does not influence TrpRS levels. It can be theorized that an immuno-suppressant effect conducted by IFN-γ explains this phenomenon, which is enhanced when IL-4 or IL-13 are linked in this formula. In inflammation situations, IFN-γ signalizes to enhance TRP storage by increasing TrpRS, preventing TRP deficiencies caused by IDO enhancement [119]. Nevertheless, in situations when IFN-γ is linked with the anti-inflammatory cytokines IL-4 and IL-13, there is an immune suppressor effect by TrpRS downregulation, reducing TRP storage and protein synthesis, finally reducing immune cell activation, preventing auto-immune

attack [118]. Janus kinase-signal transducer and activator of transcription (JAK-STAT) is a pathway that acts in the cell through IFN-γ. IFN-γ links to the JAK-STAT-1 receptor to initiate the cascade that culminates in DNA activating and transcription, generating its effects on cells [120]. In mouse microglial cells, the IFN-γ effect on IDO and TrpRS was mediated by JAK-STAT-1. The effects of IL-4 and IL-13 were not mediated by their JAK-STAT-6 traditional pathway [118].

A difference in enzyme production of the KP was observed according to the cell type after stimulation. After stimulating macrophages and microglia cells, Guillemin et al. [19] found a production of QA 20-fold greater in macrophages. This difference appears to be related to lower expression of IDO, KYNU, and KYN hydroxylase in microglia [19]. It was found that levels of QA increased in CSF of patients with depressive symptoms [69]. Then, in situations of neuroinflammation, it can suggest that the invasion of macrophages results in a more significant increase of QA, which may culminate in depressive symptoms.

## 8.8 Aging, MDD, and Kynurenine Pathway

Two critical factors are involved in aging processing. First, TRP decreases in many tissues, including the brain; second, it increases inflammatory cytokines, characterizing the senescence-associated secretory phenotype (SASP). Braidy et al. [50] found TRP levels in the brain, liver, and kidney of rats, decreasing over the aging process (3, 12, and 24 months of age). On the contrary, the KYN, QA, KYNA, IDO, TDO, and KAT increase in the aging brain (although the levels in the kidney and liver were irregular). The supposed consequences of these finds in the brain may be a 5-HT depletion from the low TRP levels, causing disorders like MDD. Another possibility is the depletion of NAD+, one of the final products of KP, which acts as a substrate for cellular energy production. Still, QA is relatively enhanced. Then, the KYNA increase may exert some protective influence for excitotoxicity exerted by QA on NMDA receptors [50]. On the other hand, a study with Alzheimer's disease (AD) subjects showed that the AD group had lower levels of KYNA than the control group. Maybe, the relative lower KYNA levels in AD can contribute to lower NMDA receptor activation, leading to memory loss related to AD [121].

Inflammatory cytokines can enhance the levels of QA by increasing the IDO cellular expression. It was recently found in untreated MDD patients with poor associative memory, a higher QA/KYNA rate. Probably the QA excitotoxic effect from NMDA receptor activation without the KYNA antagonism causes damage in memory [122]. In this regard, a study with elderly subjects (70–72 years) submitted to questionnaires to measure cognitive function found an association between poor cognitive status and high KP activation. KP activation was measured by the KIN/TRP ratio [123].

Neurodegenerative diseases like AD, Parkinson's disease (PD), and Huntington's disease (HD) are linked with MDD in terms of pathophysiological mechanisms and high prevalence of MDD in these diseases [124–126]. In elderly patients with AD and PD, the blood KYN/TRP is increased [56, 127, 128]. Increased KYN/TRP also

was found in blood samples of HD patients. KYN/TRP is increased too in CSF of PD patients [56].

Then, conditions like neuroinflammation and neurotoxicity linked with MDD, neurodegenerative diseases, and activation of the KP can permeate all these conditions, forming a complex link between the neuronal aggression mechanisms related to these conditions.

## 8.9 Gut Microbiota-Brain Axis and Kynurenine Pathway and MDD

The connection between the intestinal microbiota and the brain is called the gut-brain axis (GBA). Neuronal, endocrine, and immunological pathways establish these connections. Given this, GBA can be involved in the neurophysiology and neuropathology of various diseases such as PD, AD, MDD, and autism spectrum disorder (ASD) [129–131].

The intestinal microbiota is composed of many microbes, generally considered commensal bacteria. These organisms serve multiple purposes in the human body and exist in symbiosis with the host. Their prominent role is involved in the digestion and conversion of food materials into many useful substrates, but they also play a dynamic role in several biological functions, including strengthening the gastrointestinal epithelial barrier and resistance to pathogen invasion, promoting nutrient absorption and regulation of the functionality and maturation of the host's neuroimmune system [132, 133]. However, the microbiota is influenced by different external stimuli. Several animal studies suggest that maternal separation, containment conditions, crowding, heat, and acoustic stress alter the composition of the gut microbiota. Different forms of stress can affect the GBA [129, 134–136]. These factors can cause an imbalance between pathogenic and beneficial bacteria, stimulating the process called dysbiosis [141, 148].

Dysbiosis can alter the permeability of the intestinal barrier, and, consequently, bacteria and their metabolic products can cross to the periphery and activate the immune response [149]. Dysbiosis can increase inflammatory cytokines, and bacterial metabolites can alter the intestine and the BBB permeability [151, 152, 155]. With the disruption of the BBB and pro-inflammatory cytokines gaining access to the CNS, the peripheral QA, which under physiological conditions cannot overcome the BBB protection, reaches the CNS [142]. Consequently, a neuroinflammatory process occurs [140, 144].

Adult rats with a standard diet were subjected to a microbiome depletion paradigm followed by adoptive transfer of cecal plus colonic contents collected from donor mice fed either to high-fat diet (HFD) or control diet (CD). Mice who received the HFD microbiota showed increased anxiety and significant and selective interruptions in exploratory, cognitive (memory), and stereotypical behavior compared to those with the control diet microbiota. Mice with HFD microbiota exhibited a significant decrease in occludin expression in the jejunum and colon and claudin-3 in the colon. As these intestinal permeability markers decrease, inflammation

markers, such as inducible nitric oxide synthase and phosphorylation of the nuclear factor-kappa B subunit p65, increase in the colon of HFD mice. These results indicate increased inflammation and intestinal permeability in mice with HFD microbiota. In the analysis of the association between markers of inflammation, cerebrovascular integrity, and synaptic density in tissue homogenates prepared from the medial prefrontal cortex, an increase in the expression of microgliosis was perceived, through the expression of ionized calcium-binding adapter molecule 1 (Iba1), of toll-like receptor (TLR)-2, TLR4, and matrix metalloproteinase (MMP)-9, while endothelial tight junction proteins (zonulae occludents protein (ZO)-1 and claudin-5) and phosphorylated synapsin-1 (P-Synap) were decreased in HFD mice. These results contribute to other suggestions that alterations in the microbiota can increase neuroinflammation and interrupt cerebrovascular homeostasis [139].

Despite some studies linking GBA with neuroinflammatory and behavioral changes, the literature lacks research on the involvement of KP and the interaction of GBA and KP in MDD. This shortage is a significant gap to be considered in new studies.

## 8.10 Kynurenine Pathway and Physical Exercise and MDD

Among the risk factors for developing MDD are obesity and a sedentary lifestyle. As a result, regular physical exercise can help reduce the risk of developing the disease, in addition to helping to control when depression is established. The practice of physical exercise has benefits on mental health, in addition to causing preventive and therapeutic benefits related to psychiatric disorders. A cross-sectional survey supported the argument that physical exercise is a protective factor in the development of MDD, emphasizing a higher risk of developing the disorder in individuals with higher body mass index (BMI) and worse health, factors that may be related to a sedentary lifestyle [154].

Physical exercise is a strategy that has been used as a therapeutic adjuvant in MDD and has the potential to promote neuroplasticity and improve symptoms in both moderate depression and more severe conditions [143] in both human MDD and animal protocols [146]. Physical exercise can be a therapeutic strategy for patients' refractory to classic antidepressant treatment. Aerobic exercise, adjuvant to pharmacological therapy in treatment-resistant individuals, promoted decreasing depressive symptoms in 26% of individuals [150].

Recent studies provide evidence that among countless mechanisms underlying the effects of exercise in MDD is the modulation of the immune system, promoting neuroprotective benefits [153].

Noteworthy, in older men, the practice of physical exercise increased in skeletal muscle the transcription factors gene expression related to the KP, despite not having altered the plasma levels of KYN [138]. These results suggest that physical exercise can promote epigenetic changes in the KP. From this angle, it is crucial to highlight the peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC 1α)

gene, which is one of the genes regulated by physical exercise and that regulates the transcriptional coactivator of peroxisome-proliferator-activated γ-receptor (PPAR-γ) coactivator 1α. This transcriptional factor participates as a potent activator in mitochondrial biogenesis and oxidative metabolism, promoting increased mitochondrial density and myofibrillar proteins in muscle fibers [145, 156, 157]. Without neglecting several mechanisms through which PGC 1α can promote neuroprotection and antidepressant effect [146], KP is a relevant underlying mechanism [137].

IL-6 is known as a pro-inflammatory cytokine with altered activity in patients with MDD and related to the KP, considering that pro-inflammatory cytokines potentiate the metabolization of TRP to the KP. IL-6 levels in patients with MDD decreased with the practice of physical exercise concurrently with the reduction in the severity of depression, indicating a positive effect of physical exercise in controlling the alteration of the KP [156].

Emotionally impulsive individuals that practiced high-intensity interval training (HIIT) 3× a week for 8 weeks presented reduced blood levels of IL-6 and KP neurotoxic branch activity. In addition, HIIT contributed to controlling impulsivity, while the control group only showed positive results related to emotion. Improved impulsivity was associated with decreased IL-6 levels and increased KP neuroprotective substances, identified by KYNA/QA and KYN/QA [147].

Considering that physical exercise is related to beneficial results in MDD and many chronic diseases whose conditions involve inflammation or chronic hyper inflammation [146], it is crucial to invest in more research with protocols involving KP and other mechanisms underlying MDD.

## 8.11 Conclusion and Future Directions

This chapter sought to understand and discuss the results of translational and human research whose protocols have assessed the role of KP in the pathophysiology of MDD. Studies involving KP, or phenomena that somehow underlie or interact with the pathway, were considered. The choice was to understand the morphofunctional, metabolic, neuroprotective, and neurotoxic mechanisms and the underlying physiological processes.

Some studies have shown that inflammation is a process that is related to MDD, at least in some subtypes, in which anhedonia and reduced energy are more intense symptoms and are often associated with the severity of the disorder. In this preamble, some authors suggest that these symptoms are more related to inflammation and an imbalance, increasing neurotoxic TRP catabolites [96]. For example, KYNA and KYNA/QA ratio had a more pronounced reduction in MDD with psychotic features, which are usually more pronounced in more severe conditions of the disorder [98].

It is also essential to highlight the studies, which show that a shift in KP metabolism towards an increase in neuroprotective metabolites can protect against suicidal behavior in severe MDD. In this sense, new studies looking for metabolites or mechanisms will enable scientific advances on pharmacological targets or

therapeutic strategies aimed at the disorder's biological phenomena, which involve inflammation and KP.

Volume reduction with dendritic atrophy and neuronal loss has been observed in MDD, especially in more severe conditions that are refractory to available treatments. Some studies have shown an association between inflammation and mechanisms activated by KP with impairments in neuronal plasticity. Thus, studies aimed at the role of KP in brain morphophysiology are extremely relevant. Furthermore, study protocols with humans and animal models that can assess epigenetic alterations associated with polymorphisms in KP and brain morphology can provide the elucidation of markers and mechanisms as targets or therapeutic strategies.

Still considering advancing in investigations and possibilities of targets or more effective therapeutic strategies, it is essential to highlight the results from ECT and physical exercise protocols, pointing to the elucidation of molecular markers in the interaction of pathophysiological mechanisms.

## References

1. Organization WH. Depression and other common mental disorders: global health estimates. Geneva: World Health Organization; 2017. p. 1–24. Available from https://apps.who.int/iris/bitstream/handle/10665/254610/WHO-MSD-MER-2017.2-eng.pdf
2. Solek P, Koszla O, Mytych J, Badura J, Chelminiak Z, Cuprys M, et al. Neuronal life or death linked to depression treatment: the interplay between drugs and their stress-related outcomes relate to single or combined drug therapies. Apoptosis. 2019;24(9-10):773–84. https://doi.org/10.1007/s10495-019-01557-5.
3. Souza-Monteiro JR, Arrifano GPF, Queiroz AIDG, Mello BSF, Custodio CS, Macedo DS, et al. Antidepressant and antiaging effects of açaí (Euterpe oleracea Mart.) in mice. Oxidative Med Cell Longev. 2019;2019:1–16. https://doi.org/10.1155/2019/3614960.
4. Underwood MD, Kassir SA, Bakalian MJ, Galfalvy H, Dwork AJ, Mann JJ, et al. Serotonin receptors and suicide, major depression, alcohol use disorder and reported early life adversity. Transl Psychiatry. 2018;8(1):279. https://doi.org/10.1038/s41398-018-0309-1.
5. Tao X, Yan M, Wang L, Zhou Y, Wang Z, Xia T, et al. Homeostasis imbalance of microglia and astrocytes leads to alteration in the metabolites of the kynurenine pathway in LPS-induced depressive-like mice. Int J Mol Sci. 2020;21(4):1460. https://doi.org/10.3390/ijms21041460.
6. Heine W, Radke M, Wutzke KD. The significance of tryptophan in human nutrition. Amino Acids. 1995;9(3):91–205. https://doi.org/10.1007/BF00805951.
7. Colle R, Masson P, Verstuyft C, Fève B, Werner E, Boursier-Neyret C, et al. Peripheral tryptophan, serotonin, kynurenine, and their metabolites in major depression: a case–control study. Psychiatry Clin Neurosci. 2020;74(2):112–7. https://doi.org/10.1111/pcn.12944.
8. Mori Y, Mouri A, Kunisawa K, Hirakawa M, Kubota H, Kosuge A, et al. Kynurenine 3-monooxygenase deficiency induces depression-like behavior via enhanced antagonism of α7 nicotinic acetylcholine receptors by kynurenic acid. Behav Brain Res. 2021;405:113191. https://doi.org/10.1016/j.bbr.2021.113191.
9. Miranda AF, Boegman RJ, Beninger RJ, Jhamandas K. Protection against quinolinic acid-mediated excitotoxicity in nigrostriatal dopaminergic neurons by endogenous kynurenic acid. Neuroscience. 1997;78(4):967–75. https://doi.org/10.1016/S0306-4522(96)00655-0.
10. Kindler J, Lim CK, Weickert CS, Boerrigter D, Galletly C, Liu D, et al. Dysregulation of kynurenine metabolism is related to proinflammatory cytokines, attention, and prefrontal cortex volume in schizophrenia. Mol Psychiatry. 2020;25(11):2860–72. https://doi.org/10.1038/s41380-019-0401-9.

11. Liu W, Ge T, Leng Y, Pan Z, Fan J, Yang W, et al. The role of neural plasticity in depression: from hippocampus to prefrontal cortex. Neural Plast. 2017;2017:1–11. https://doi.org/10.1155/2017/6871089.
12. Martín-Hernández D, Tendilla-Beltrán H, Madrigal JLM, García-Bueno B, Leza JC, Caso JR. Chronic mild stress alters kynurenine pathways changing the glutamate neurotransmission in frontal cortex of rats. Mol Neurobiol. 2019;56(1):490–501. https://doi.org/10.1007/s12035-018-1096-7.
13. Bryleva EY, Brundin L. Suicidality and activation of the kynurenine pathway of tryptophan metabolism. Curr Top Behav Neurosci. 2017;31:269–84. https://doi.org/10.1007/7854_2016_5.
14. Maes M, Leonard BE, Myint AM, Kubera M, Verkerk R. The new "5-HT" hypothesis of depression: cell-mediated immune activation induces indoleamine 2,3-dioxygenase, which leads to lower plasma tryptophan and an increased synthesis of detrimental tryptophan catabolites (TRYCATs), both of which contribute to the onset of depression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2011;35(3):702–21. https://doi.org/10.1016/j.pnpbp.2010.12.017.
15. O'Connor JC, André C, Wang Y, Lawson MA, Szegedi SS, Lestage J, et al. Interferon-γ and tumor necrosis factor-α mediate the upregulation of indoleamine 2,3-dioxygenase and the induction of depressive-like behavior in mice in response to bacillus calmette-guérin. J Neurosci. 2009;29(13):4200–9. https://doi.org/10.1523/JNEUROSCI.5032-08.2009.
16. Won E, Kim Y-K. Stress, the autonomic nervous system, and the immune-kynurenine pathway in the etiology of depression. Curr Neuropharmacol. 2016;14(7):665–73. https://doi.org/10.2174/1570159X14666151208113006.
17. Han Q, Robinson H, Li J. Biochemical identification and crystal structure of kynurenine formamidase from Drosophila melanogaster. Biochem J. 2012;446(2):253–60. https://doi.org/10.1042/BJ20120416.
18. Heyes MP, Saito K, Crowley JS, Davis LE, Demitrack MA, Der M, et al. Quinolinic acid and kynurenine pathway metabolism in inflammatory and non-inflammatory neurological disease. Brain. 1992;115(5):1249–73. https://doi.org/10.1093/brain/115.5.1249.
19. Guillemin GJ, Smith DG, Smythe GA, Armati PJ, Brew GJ. Expression of the kynurenine pathway enzymes in human microglia and macrophages. Adv Exp Med Biol. 2003;527:105–12. https://doi.org/10.1007/978-1-4615-0135-0_12.
20. Guillemin GJ, Kerr SJ, Smythe GA, Smith DG, Kapoor V, Armati PJ, et al. Kynurenine pathway metabolism in human astrocytes: a paradox for neuronal protection. J Neurochem. 2001;78(4):842–53. https://doi.org/10.1046/j.1471-4159.2001.00498.x.
21. Guidetti P, Amori L, Sapko MT, Okuno E, Schwarcz R. Mitochondrial aspartate aminotransferase: a third kynurenate-producing enzyme in the mammalian brain. J Neurochem. 2007;102(1):103–11. https://doi.org/10.1111/j.1471-4159.2007.04556.x.
22. Han Q, Cai T, Tagle DA, Li J. Structure, expression, and function of kynurenine aminotransferases in human and rodent brains. Cell Mol Life Sci. 2010;67(3):353–68. https://doi.org/10.1007/s00018-009-0166-4.
23. Arteaga-Henriquez G, Burger B, Weidinger E, Grosse L, Moll N, Schuetze G, et al. Activation and deactivation steps in the tryptophan breakdown pathway in major depressive disorder: a link to the monocyte inflammatory state of patients. Prog Neuro-Psychopharmacol Biol Psychiatry. 2021;107:110226. https://doi.org/10.1016/j.pnpbp.2020.110226.
24. Stone TW, Darlington LG. Endogenous kynurenines as targets for drug discovery and development. Nat Rev Drug Discov. 2002;1(8):609–20. https://doi.org/10.1038/nrd870.
25. Heisler JM, O'Connor JC. Indoleamine 2,3-dioxygenase-dependent neurotoxic kynurenine metabolism mediates inflammation-induced deficit in recognition memory. Brain Behav Immun. 2015;50:115–24. https://doi.org/10.1016/j.bbi.2015.06.022.
26. Moroni F. Tryptophan metabolism and brain function: focus on kynurenine and other indole metabolites. Eur J Pharmacol. 1999;375(1–3):87–100. https://doi.org/10.1016/S0014-2999(99)00196-X.

27. Perkins MN, Stone TW. An iontophoretic investigation of the actions of convulsant kynurenines and their interaction with the endogenous excitant quinolinic acid. Brain Res. 1982;247(1):184–7. https://doi.org/10.1016/0006-8993(82)91048-4.
28. Parsons CG, Danysz W, Quack G, Hartmann S, Lorenz B, Wollenburg C, et al. Novel systemically active antagonists of the glycine site of the N-methyl-D-aspartate receptor: electrophysiological, biochemical and behavioral characterization. J Pharmacol Exp Ther. 1997;283(3):1264–75. Available from: http://www.ncbi.nlm.nih.gov/pubmed/9400002
29. Hilmas C, Pereira EFR, Alkondon M, Rassoulpour A, Schwarcz R, Albuquerque EX. The brain metabolite kynurenic acid inhibits α7 nicotinic receptor activity and increases non-α7 nicotinic receptor expression: physiopathological implications. J Neurosci. 2001;21(19):7463–73. https://doi.org/10.1523/JNEUROSCI.21-19-07463.2001.
30. Carpenedo R, Pittaluga A, Cozzi A, Attucci S, Galli A, Raiteri M, et al. Presynaptic kynurenate-sensitive receptors inhibit glutamate release. Eur J Neurosci. 2001;13(11):2141–7. https://doi.org/10.1046/j.0953-816X.2001.01592.x.
31. Grilli M, Raiteri L, Patti L, Parodi M, Robino F, Raiteri M, et al. Modulation of the function of presynaptic α7 and non-α7 nicotinic receptors by the tryptophan metabolites, 5-hydroxyindole and kynurenate in mouse brain. Br J Pharmacol. 2006;149(6):724–32. https://doi.org/10.1038/sj.bjp.0706914.
32. Rassoulpour A, Wu HQ, Ferre S, Schwarcz R. Nanomolar concentrations of kynurenic acid reduce extracellular dopamine levels in the striatum. J Neurochem. 2005;93(3):762–5. https://doi.org/10.1111/j.1471-4159.2005.03134.x.
33. Zmarowski A, Wu HQ, Brooks JM, Potter MC, Pellicciari R, Schwarcz R, et al. Astrocyte-derived kynurenic acid modulates basal and evoked cortical acetylcholine release. Eur J Neurosci. 2009;29(3):529–38. https://doi.org/10.1111/j.1460-9568.2008.06594.x.
34. Chiarugi A, Carpenedo R, Molina MT, Mattoli L, Pellicciari R, Moroni F. Comparison of the neurochemical and behavioral effects resulting from the inhibition of kynurenine hydroxylase and/or kynureninase. J Neurochem. 2002;65(3):1176–83. https://doi.org/10.1046/j.1471-4159.1995.65031176.x.
35. Huang Y-S, Ogbechi J, Clanchy FI, Williams RO, Stone TW. IDO and kynurenine metabolites in peripheral and CNS disorders. Front Immunol. 2020;11:388. https://doi.org/10.3389/fimmu.2020.00388.
36. Ramírez-Ortega D, Ramiro-Salazar A, González-Esquivel D, Ríos C, Pineda B, Pérez De La Cruz V. 3-Hydroxykynurenine and 3-hydroxyanthranilic acid enhance the toxicity induced by copper in rat astrocyte culture. Oxidative Med Cell Longev. 2017;2017:1–12. https://doi.org/10.1155/2017/2371895.
37. Stone TW. Endogenous neurotoxins from tryptophan. Toxicon. 2001;39(1):61–73. https://doi.org/10.1016/S0041-0101(00)00156-2.
38. Kawai J, Okuno E, Kido R. Organ distribution of rat kynureninase and changes of its activity during development. Enzyme. 1988;39(4):181–9. https://doi.org/10.1159/000469117.
39. Goldstein LE, Leopold MC, Huang X, Atwood CS, Saunders AJ, Hartshorn M, et al. 3-Hydroxykynurenine and 3-hydroxyanthranilic acid generate hydrogen peroxide and promote A-crystallin cross-linking by metal ion reduction. Biochemistry. 2000;39(24):7266–75. https://doi.org/10.1021/bi992997s.
40. Katsyuba E, Mottis A, Zietak M, De Franco F, van der Velpen V, Gariani K, et al. De novo NAD+ synthesis enhances mitochondrial function and improves health. Nature. 2018;563(7731):354–9. https://doi.org/10.1038/s41586-018-0645-6.
41. Schwarcz R, Bruno JP, Muchowski PJ, Wu HQ. Kynurenines in the mammalian brain: when physiology meets pathology. Nat Rev Neurosci. 2012;13(7):465–77. https://doi.org/10.1038/nrn3257.
42. Wang Q, Liu D, Song P, Zou MH. Tryptophan-kynurenine pathway is dysregulated in inflammation, and immune activation. Front Biosci Landmark. 2015;20(7):1116–43. https://doi.org/10.2741/4363.

43. La Cruz VP, Carrillo-Mora P, Santamaría A. Quinolinic acid, an endogenous molecule combining excitotoxicity, oxidative stress and other toxic mechanisms. Int J Tryptophan Res. 2013;5(1):1–8. https://doi.org/10.4137/IJTR.S8158.
44. Stone TW, Perkins MN. Quinolinic acid: a potent endogenous excitant at amino acid receptors in CNS. Eur J Pharmacol. 1981;72(4):411–2. https://doi.org/10.1016/0014-2999(81)90587-2.
45. Tavares RG, Tasca CI, Santos CES, Alves LB, Porciúncula LO, Emanuelli T, et al. Quinolinic acid stimulates synaptosomal glutamate release and inhibits glutamate uptake into astrocytes. Neurochem Int. 2002;40(7):621–7. https://doi.org/10.1016/S0197-0186(01)00133-4.
46. Dantzer R. Role of the kynurenine metabolism pathway in inflammation-induced depression: preclinical approaches. Curr Top Behav Neurosci. 2017;31:117–38. https://doi.org/10.1007/7854_2016_6.
47. Myint AM, Kim YK, Verkerk R, Scharpé S, Steinbusch H, Leonard B. Kynurenine pathway in major depression: evidence of impaired neuroprotection. J Affect Disord. 2007;98(1–2):143–51. https://doi.org/10.1016/j.jad.2006.07.013.
48. Sharma A, Kazim SF, Larson CS, Ramakrishnan A, Gray JD, McEwen BS, et al. Divergent roles of astrocytic versus neuronal EAAT2 deficiency on cognition and overlap with aging and Alzheimer's molecular signatures. Proc Natl Acad Sci. 2019;116(43):21800–11. https://doi.org/10.1073/pnas.1903566116.
49. Mattson MP. Excitotoxicity. In: Stress: physiology, biochemistry, and pathology handbook of stress series, vol. 3. Amsterdam: Elsevier; 2019. p. 125–34. https://doi.org/10.1016/B978-0-12-813146-6.00011-4.
50. Braidy N, Guillemin GJ, Mansour H, Chan-Ling T, Grant R. Changes in kynurenine pathway metabolism in the brain, liver and kidney of aged female Wistar rats. FEBS J. 2011;278(22):4425–34. https://doi.org/10.1111/j.1742-4658.2011.08366.x.
51. Bouchard VJ, Rouleau M, Poirier GG. PARP-1, a determinant of cell survival in response to DNA damage. Exp Hematol. 2003;31(6):446–54. https://doi.org/10.1016/S0301-472X(03)00083-3.
52. Erdélyi K, Bakondi E, Gergely P, Szabó C, Virág L. Pathophysiologic role of oxidative stress-induced poly(ADP-ribose) polymerase-1 activation: focus on cell death and transcriptional regulation. Cell Mol Life Sci. 2005;62(7–8):751–9. https://doi.org/10.1007/s00018-004-4506-0.
53. Haroon E, Welle JR, Woolwine BJ, Goldsmith DR, Baer W, Patel T, et al. Associations among peripheral and central kynurenine pathway metabolites and inflammation in depression. Neuropsychopharmacology. 2020;45(6):998–1007. https://doi.org/10.1038/s41386-020-0607-1.
54. Imbeault S, Goiny M, Liu X, Erhardt S. Effects of IDO1 and TDO2 inhibition on cognitive deficits and anxiety following LPS-induced neuroinflammation. Acta Neuropsychiatr. 2020;32(1):43–53. https://doi.org/10.1017/neu.2019.44.
55. Yan J, Kuzhiumparambil U, Bandodkar A, Bandodkar S, Dale RC, Fu S. Cerebrospinal fluid metabolites in tryptophan-kynurenine and nitric oxide pathways: biomarkers for acute neuroinflammation. Dev Med Child Neurol. 2021;63(5):552–9. https://doi.org/10.1111/dmcn.14774.
56. Widner B, Leblhuber F, Fuchs D. Increased neopterin production and tryptophan degradation in advanced Parkinson's disease. J Neural Transm. 2002;109(2):181–9. https://doi.org/10.1007/s007020200014.
57. Yan J, Kuzhiumparambil U, Bandodkar S, Dale RC, Fu S. Cerebrospinal fluid metabolomics: detection of neuroinflammation in human central nervous system disease. Clin Transl Immunol. 2021;10(8):1318. https://doi.org/10.1002/cti2.1318.
58. Quist-Paulsen E, Aukrust P, Kran A-MB, Dunlop O, Ormaasen V, Stiksrud B, et al. High neopterin and IP-10 levels in cerebrospinal fluid are associated with neurotoxic tryptophan metabolites in acute central nervous system infections. J Neuroinflammation. 2018;15(1):327. https://doi.org/10.1186/s12974-018-1366-3.

59. Kruse JL, Cho JHJ, Olmstead R, Hwang L, Faull K, Eisenberger NI, et al. Kynurenine metabolism and inflammation-induced depressed mood: a human experimental study. Psychoneuroendocrinology. 2019;109:104371. https://doi.org/10.1016/j.psyneuen.2019.104371.
60. Murata S, Murphy M, Hoppensteadt D, Fareed J, Welborn A, Halaris A. Effects of adjunctive inflammatory modulation on IL-1β in treatment resistant bipolar depression. Brain Behav Immun. 2020;87:369–76. https://doi.org/10.1016/j.bbi.2020.01.004.
61. Verheyen N, Meinitzer A, Grübler MR, Ablasser K, Kolesnik E, Fahrleitner-Pammer A, et al. Low-grade inflammation and tryptophan-kynurenine pathway activation are associated with adverse cardiac remodeling in primary hyperparathyroidism: the EPATH trial. Clin Chem Lab Med. 2017;55(7):1034–42. https://doi.org/10.1515/cclm-2016-1159.
62. Fuertig R, Azzinnari D, Bergamini G, Cathomas F, Sigrist H, Seifritz E, et al. Mouse chronic social stress increases blood and brain kynurenine pathway activity and fear behaviour: both effects are reversed by inhibition of indoleamine 2,3-dioxygenase. Brain Behav Immun. 2016;54:59–72. https://doi.org/10.1016/j.bbi.2015.12.020.
63. Kendler KS, Karkowski LM, Prescott CA. Causal relationship between stressful life events and the onset of major depression. Am J Psychiatry. 1999;156(6):837–41. https://doi.org/10.1176/ajp.156.6.837.
64. Bierhaus A, Wolf J, Andrassy M, Rohleder N, Humpert PM, Petrov D, et al. A mechanism converting psychosocial stress into mononuclear cell activation. Proc Natl Acad Sci U S A. 2003;100(4):1920–5. https://doi.org/10.1073/pnas.0438019100.
65. Rohleder N. Stimulation of systemic low-grade inflammation by psychosocial stress. Psychosom Med. 2014;76(3):181–9. https://doi.org/10.1097/PSY.0000000000000049.
66. Wohleb ES, Hanke ML, Corona AW, Powell ND, Stiner LM, Bailey MT, et al. β-Adrenergic receptor antagonism prevents anxiety-like behavior and microglial reactivity induced by repeated social defeat. J Neurosci. 2011;31(17):6277–88. https://doi.org/10.1523/JNEUROSCI.0450-11.2011.
67. Wohleb ES, Powell ND, Godbout JP, Sheridan JF. Stress-induced recruitment of bone marrow-derived monocytes to the brain promotes anxiety-like behavior. J Neurosci. 2013;33(34):13820–33. https://doi.org/10.1523/JNEUROSCI.1671-13.2013.
68. Myint AM. Kynurenines: from the perspective of major psychiatric disorders. FEBS J. 2012;279(8):1375–85. https://doi.org/10.1111/j.1742-4658.2012.08551.x.
69. Raison CL, Dantzer R, Kelley KW, Lawson MA, Woolwine BJ, Vogt G, et al. CSF concentrations of brain tryptophan and kynurenines during immune stimulation with IFN-α: relationship to CNS immune responses and depression. Mol Psychiatry. 2010;15(4):393–403. https://doi.org/10.1038/mp.2009.116.
70. Bay-Richter C, Linderholm KR, Lim CK, Samuelsson M, Träskman-Bendz L, Guillemin GJ, et al. A role for inflammatory metabolites as modulators of the glutamate N-methyl-d-aspartate receptor in depression and suicidality. Brain Behav Immun. 2015;43:110–7. https://doi.org/10.1016/j.bbi.2014.07.012.
71. Kim H, Chen L, Lim G, Sung B, Wang S, McCabe MF, et al. Brain indoleamine 2,3-dioxygenase contributes to the comorbidity of pain and depression. J Clin Invest. 2012;122(8):2940–54. https://doi.org/10.1172/JCI61884.
72. Réus GZ, Jansen K, Titus S, Carvalho AF, Gabbay V, Quevedo J. Kynurenine pathway dysfunction in the pathophysiology and treatment of depression: Evidences from animal and human studies. J Psychiatr Res. 2015;68:316–28. https://doi.org/10.1016/j.jpsychires.2015.05.007.
73. Savitz J, Drevets WC, Smith CM, Victor TA, Wurfel BE, Bellgowan PSF, et al. Putative neuroprotective and neurotoxic kynurenine pathway metabolites are associated with hippocampal and amygdalar volumes in subjects with major depressive disorder. Neuropsychopharmacology. 2015;40(2):463–71. https://doi.org/10.1038/npp.2014.194.
74. Steiner J, Walter M, Gos T, Guillemin GJ, Bernstein HG, Sarnyai Z, et al. Severe depression is associated with increased microglial quinolinic acid in subregions of the anterior cingulate

gyrus: evidence for an immune-modulated glutamatergic neurotransmission? J Neuroinflammation. 2011;8(1):94. https://doi.org/10.1186/1742-2094-8-94.
75. Sublette ME, Galfalvy HC, Fuchs D, Lapidus M, Grunebaum MF, Oquendo MA, et al. Plasma kynurenine levels are elevated in suicide attempters with major depressive disorder. Brain Behav Immun. 2011;25(6):1272–8. https://doi.org/10.1016/j.bbi.2011.05.002.
76. Tsigos C, Chrousos GP. Hypothalamic–pituitary–adrenal axis, neuroendocrine factors and stress. J Psychosom Res. 2002;53(4):865–71. https://doi.org/10.1016/S0022-3999(02)00429-4.
77. Badawy AAB. Kynurenine pathway of tryptophan metabolism: regulatory and functional aspects. Int J Tryptophan Res. 2017;10(1):117864691769193. https://doi.org/10.1177/1178646917691938.
78. Nakamura T, Niimi S, Nawa K, Noda C, Ichihara A, Takagi Y, et al. Multihormonal regulation of transcription of the tryptophan 2,3-dioxygenase gene in primary cultures of adult rat hepatocytes with special reference to the presence of a transcriptional protein mediating the action of glucocorticoids. J Biol Chem. 1987;262(2):727–33. https://doi.org/10.1016/s0021-9258(19)75845-1.
79. Maes M, Jacobs MP, Suy E, Vandewoude M, Minner B, Raus J. Effects of dexamethasone on the availability of l-tryptophan and on the insulin and FFA concentrations in unipolar depressed patients. Biol Psychiatry. 1990;27(8):854–62. https://doi.org/10.1016/0006-3223(90)90466-F.
80. Ogawa S, Fujii T, Koga N, Hori H, Teraishi T, Hattori K, et al. Plasma l-tryptophan concentration in major depressive disorder: new data and meta-analysis. J Clin Psychiatry. 2014;75(9):e906–15. https://doi.org/10.4088/JCP.13r08908.
81. Cohen S, Janicki-Deverts D, Doyle WJ, Miller GE, Frank E, Rabin BS, et al. Chronic stress, glucocorticoid receptor resistance, inflammation, and disease risk. Proc Natl Acad Sci U S A. 2012;109(16):5995–9. https://doi.org/10.1073/pnas.1118355109.
82. Sorgdrager FJH, Doornbos B, Penninx BWJH, de Jonge P, Kema IP. The association between the hypothalamic pituitary adrenal axis and tryptophan metabolism in persons with recurrent major depressive disorder and healthy controls. J Affect Disord. 2017;222:32–9. https://doi.org/10.1016/j.jad.2017.06.052.
83. Firk C, Markus CR. Mood and cortisol responses following tryptophan-rich hydrolyzed protein and acute stress in healthy subjects with high and low cognitive reactivity to depression. Clin Nutr. 2009;28(3):266–71. https://doi.org/10.1016/j.clnu.2009.03.002.
84. Messaoud A, Mensi R, Douki W, Neffati F, Najjar MF, Gobbi G, et al. Reduced peripheral availability of tryptophan and increased activation of the kynurenine pathway and cortisol correlate with major depression and suicide. World J Biol Psychiatry. 2019;20(9):703–11. https://doi.org/10.1080/15622975.2018.1468031.
85. Dornbierer DA, Boxler M, Voegel CD, Stucky B, Steuer AE, Binz TM, et al. Nocturnal gamma-hydroxybutyrate reduces cortisol-awakening response and morning kynurenine pathway metabolites in healthy volunteers. Int J Neuropsychopharmacol. 2019;22(10):631–9. https://doi.org/10.1093/ijnp/pyz047.
86. Robinson CM, Hale PT, Carlin JM. The role of IFN-γ and TNF-α-responsive regulatory elements in the synergistic induction of indoleamine dioxygenase. J Interf Cytokine Res. 2005;25(1):20–30. https://doi.org/10.1089/jir.2005.25.20.
87. Marx W, McGuinness AJ, Rocks T, Ruusunen A, Cleminson J, Walker AJ, et al. The kynurenine pathway in major depressive disorder, bipolar disorder, and schizophrenia: a meta-analysis of 101 studies. Mol Psychiatry. 2021;26(8):4158–78. https://doi.org/10.1038/s41380-020-00951-9.
88. Bradley KAL, Case JAC, Khan O, Ricart T, Hanna A, Alonso CM, et al. The role of the kynurenine pathway in suicidality in adolescent major depressive disorder. Psychiatry Res. 2015;227(2–3):206–12. https://doi.org/10.1016/j.psychres.2015.03.031.
89. Brundin L, Sellgren CM, Lim CK, Grit J, Pålsson E, Landén M, et al. An enzyme in the kynurenine pathway that governs vulnerability to suicidal behavior by regulating

excitotoxicity and neuroinflammation. Transl Psychiatry. 2016;6(8):e865. https://doi.org/10.1038/tp.2016.133.
90. Abbott CC, Lemke NT, Gopal S, Thoma RJ, Bustillo J, Calhoun VD, et al. Electroconvulsive therapy response in major depressive disorder: a pilot functional network connectivity resting state fMRI investigation. Front Psych. 2013;4:10. https://doi.org/10.3389/fpsyt.2013.00010.
91. Schwieler L, Samuelsson M, Frye MA, Bhat M, Schuppe-Koistinen I, Jungholm O, et al. Electroconvulsive therapy suppresses the neurotoxic branch of the kynurenine pathway in treatment-resistant depressed patients. J Neuroinflammation. 2016;13(1):51. https://doi.org/10.1186/s12974-016-0517-7.
92. Achtyes E, Keaton SA, Smart LA, Burmeister AR, Heilman PL, Krzyzanowski S, et al. Inflammation and kynurenine pathway dysregulation in post-partum women with severe and suicidal depression. Brain Behav Immun. 2020;83:239–47. https://doi.org/10.1016/j.bbi.2019.10.017.
93. Han KM, Ham BJ. How inflammation affects the brain in depression: a review of functional and structural MRI studies. J Clin Neurol. 2021;17(4):503–15. https://doi.org/10.3988/jcn.2021.17.4.503.
94. Meier TB, Drevets WC, Wurfel BE, Ford BN, Morris HM, Victor TA, et al. Relationship between neurotoxic kynurenine metabolites and reductions in right medial prefrontal cortical thickness in major depressive disorder. Brain Behav Immun. 2016;53:39–48. https://doi.org/10.1016/j.bbi.2015.11.003.
95. Zhou Y, Zheng W, Liu W, Wang C, Zhan Y, Li H, et al. Cross-sectional relationship between kynurenine pathway metabolites and cognitive function in major depressive disorder. Psychoneuroendocrinology. 2019;101:72–9. https://doi.org/10.1016/j.psyneuen.2018.11.001.
96. Cho HJ, Savitz J, Dantzer R, Teague TK, Drevets WC, Irwin MR. Sleep disturbance and kynurenine metabolism in depression. J Psychosom Res. 2017;99:1–7. https://doi.org/10.1016/j.jpsychores.2017.05.016.
97. Milaneschi Y, Allers KA, Beekman ATF, Giltay EJ, Keller S, Schoevers RA, et al. The association between plasma tryptophan catabolites and depression: the role of symptom profiles and inflammation. Brain Behav Immun. 2021;97:167–75. https://doi.org/10.1016/j.bbi.2021.07.007.
98. Wurfel BE, Drevets WC, Bliss SA, McMillin JR, Suzuki H, Ford BN, et al. Serum kynurenic acid is reduced in affective psychosis. Transl Psychiatry. 2017;7(5):e1115. https://doi.org/10.1038/tp.2017.88.
99. Sakurai M, Yamamoto Y, Kanayama N, Hasegawa M, Mouri A, Takemura M, et al. Serum metabolic profiles of the tryptophan-kynurenine pathway in the high risk subjects of major depressive disorder. Sci Rep. 2020;10(1):1961. https://doi.org/10.1038/s41598-020-58806-w.
100. Laugeray A, Launay J-M, Callebert J, Surget A, Belzung C, Barone PR. Peripheral and cerebral metabolic abnormalities of the tryptophan–kynurenine pathway in a murine model of major depression. Behav Brain Res. 2010;210(1):84–91. https://doi.org/10.1016/j.bbr.2010.02.014.
101. Tashiro T, Murakami Y, Mouri A, Imamura Y, Nabeshima T, Yamamoto Y, et al. Kynurenine 3-monooxygenase is implicated in antidepressants-responsive depressive-like behaviors and monoaminergic dysfunctions. Behav Brain Res. 2017;317:279–85. https://doi.org/10.1016/j.bbr.2016.09.050.
102. Tanaka M, Bohár Z, Martos D, Telegdy G, Vécsei L. Antidepressant-like effects of kynurenic acid in a modified forced swim test. Pharmacol Rep. 2020;72(2):449–55. https://doi.org/10.1007/s43440-020-00067-5.
103. Ader R, Cohen N, Felten D. Psychoneuroimmunology: interactions between the nervous system and the immune system. Lancet. 1995;345(8942):99–103. https://doi.org/10.1016/S0140-6736(95)90066-7.
104. Gold PW, Machado-Vieira R, Pavlatou MG. Clinical and biochemical manifestations of depression: relation to the neurobiology of stress. Neural Plast. 2015;2015:1–11. https://doi.org/10.1155/2015/581976.

105. Hu MX, Lamers F, De Geus EJC, Penninx BWJH. Differential autonomic nervous system reactivity in depression and anxiety during stress depending on type of stressor. Psychosom Med. 2016;78(5):562–72. https://doi.org/10.1097/PSY.0000000000000313.
106. Cameron O. Depression increases post-Mi mortality: how? Psychosom Med. 1996;58(2): 111–2. https://doi.org/10.1097/00006842-199603000-00002.
107. Carney RM, Freedland KE, Miller GE, Jaffe AS. Depression as a risk factor for cardiac mortality and morbidity: a review of potential mechanisms. J Psychosom Res. 2002;53(4): 897–902. https://doi.org/10.1016/S0022-3999(02)00311-2.
108. Glassman AH, Shapiro PA. Depression and the course of coronary artery disease. Am J Psychiatry. 1998;155(1):4–11. https://doi.org/10.1176/ajp.155.1.4.
109. Raič M. Depression and heart diseases: leading health problems. Psychiatr Danub. 2017;29: 770–7.
110. Irwin MR. Depression and immunity: central corticotropin-releasing factor activates the autonomic nervous system and reduces natural killer cell activity. In: Stress and disease processes: perspectives in behavioral medicine. Washington: Psychology Press; 2018. p. 103–19. https://doi.org/10.4324/9781315827490-6.
111. Jones BE, Yang T-Z. The efferent projections from the reticular formation and the locus coeruleus studied by anterograde and retrograde axonal transport in the rat. J Comp Neurol. 1985;242(1):56–92. https://doi.org/10.1002/cne.902420105.
112. Lewis DI, Coote JH. Excitation and inhibition of rat sympathetic preganglionic neurones by catecholamines. Brain Res. 1990;530(2):229–34. https://doi.org/10.1016/0006-8993(90)91287-Q.
113. Streeter CC, Gerbarg PL, Saper RB, Ciraulo DA, Brown RP. Effects of yoga on the autonomic nervous system, gamma-aminobutyric-acid, and allostasis in epilepsy, depression, and post-traumatic stress disorder. Med Hypotheses. 2012;78(5):571–9. https://doi.org/10.1016/j.mehy.2012.01.021.
114. Chrousos GP, Gold PW. The concepts of stress and stress system disorders: overview of physical and behavioral homeostasis. JAMA J Am Med Assoc. 1992;267(9):1244–52. https://doi.org/10.1001/jama.1992.03480090092034.
115. Halaris A. Inflammation-associated co-morbidity between depression and cardiovascular disease. Curr Top Behav Neurosci. 2017;31:45–70. https://doi.org/10.1007/7854_2016_28.
116. O'Connor JC, Lawson MA, André C, Briley EM, Szegedi SS, Lestage J, et al. Induction of IDO by Bacille Calmette-Guérin is responsible for development of murine depressive-like behavior. J Immunol. 2009;182(5):3202–12. https://doi.org/10.4049/jimmunol.0802722.
117. Rodriguez-Zas SL, Wu C, Southey BR, O'Connor JC, Nixon SE, Garcia R, et al. Disruption of microglia histone acetylation and protein pathways in mice exhibiting inflammation-associated depression-like symptoms. Psychoneuroendocrinology. 2018;97:47–58. https://doi.org/10.1016/j.psyneuen.2018.06.024.
118. Yadav MC, Burudi EME, Alirezaei M, Flynn CC, Watry DD, Lanigan CM, et al. IFN-γ-induced IDO and WRS expression in microglia is differentially regulated by IL-4. Glia. 2007;55(13):1385–96. https://doi.org/10.1002/glia.20544.
119. Ahn YH, Oh S-C, Zhou S, Kim T-D. Tryptophanyl-tRNA synthetase as a potential therapeutic target. Int J Mol Sci. 2021;22(9):4523. https://doi.org/10.3390/ijms22094523.
120. Nguyen VT, Benveniste EN. Involvement of STAT-1 and Ets family members interferon-γ induction of CD40 transcription in microglia/macrophages. J Biol Chem. 2000;275(31): 23674–84. https://doi.org/10.1074/jbc.M002482200.
121. Hartai Z, Juhász A, Rimanóczy Á, Janáky T, Donkó T, Dux L, et al. Decreased serum and red blood cell kynurenic acid levels in Alzheimer's disease. Neurochem Int. 2007;50(2):308–13. https://doi.org/10.1016/j.neuint.2006.08.012.
122. Chirico M, Custer J, Shoyombo I, Cooper C, Meldrum S, Dantzer R, et al. Kynurenine pathway metabolites selectively associate with impaired associative memory function in depression. Brain Behav Immun Health. 2020;8:100126. https://doi.org/10.1016/j.bbih.2020.100126.

123. Solvang SEH, Nordrehaug JE, Tell GS, Nygård O, McCann A, Ueland PM, et al. The kynurenine pathway and cognitive performance in community-dwelling older adults. The Hordaland Health Study. Brain Behav Immun. 2019;75:155–62. https://doi.org/10.1016/j.bbi.2018.10.003.
124. Paulsen JS, Hoth KF, Nehl C, Stierman L. Critical periods of suicide risk in Huntington's disease. Am J Psychiatry. 2005;162(4):725–31. https://doi.org/10.1176/appi.ajp.162.4.725.
125. Reijnders JSAM, Ehrt U, Weber WEJ, Aarsland D, Leentjens AFG. A systematic review of prevalence studies of depression in Parkinson's disease. Mov Disord. 2008;23(2):183–9. https://doi.org/10.1002/mds.21803.
126. Zhao Q-F, Tan L, Wang H-F, Jiang T, Tan M-S, Tan L, et al. The prevalence of neuropsychiatric symptoms in Alzheimer's disease: systematic review and meta-analysis. J Affect Disord. 2016;190:264–71. https://doi.org/10.1016/j.jad.2015.09.069.
127. Gulaj E, Pawlak K, Bien B, Pawlak D. Kynurenine and its metabolites in Alzheimer's disease patients. Adv Med Sci. 2010;55(2):204–11. https://doi.org/10.2478/v10039-010-0023-6.
128. Widner B, Leblhuber F, Walli J, Tilz GP, Demel U, Fuchs D. Tryptophan degradation and immune activation in Alzheimer's disease. J Neural Transm. 2000;107(3):343–53. https://doi.org/10.1007/s007020050029.
129. Mayer EA, Knight R, Mazmanian SK, Cryan JF, Tillisch K. Gut microbes and the brain: paradigm shift in neuroscience. J Neurosci. 2014;34(46):15490–6. https://doi.org/10.1523/JNEUROSCI.3299-14.2014.
130. Rajanala K, Kumar N, Chamallamudi MR. Modulation of gut-brain axis by probiotics: a promising anti-depressant approach. Curr Neuropharmacol. 2021;19(7):990–1006. https://doi.org/10.2174/1570159x19666201215142520.
131. Smith LM, Parr-Brownlie LC. A neuroscience perspective of the gut theory of Parkinson's disease. Eur J Neurosci. 2019;49(6):817–23. https://doi.org/10.1111/ejn.13869.
132. Neish AS. Microbes in gastrointestinal health and disease. Gastroenterology. 2009;136(1):65–80. https://doi.org/10.1053/j.gastro.2008.10.080.
133. Sekirov I, Russell SL, Caetano M, Antunes L, Finlay BB. Gut microbiota in health and disease. Physiol Rev. 2010;90(3):859–904. https://doi.org/10.1152/physrev.00045.2009.
134. Bailey MT, Dowd SE, Galley JD, Hufnagle AR, Allen RG, Lyte M. Exposure to a social stressor alters the structure of the intestinal microbiota: Implications for stressor-induced immunomodulation. Brain Behav Immun. 2011;25(3):397–407. https://doi.org/10.1016/j.bbi.2010.10.023.
135. De Palma G, Collins SM, Bercik P, Verdu EF. The microbiota-gut-brain axis in gastrointestinal disorders: Stressed bugs, stressed brain or both? J Physiol. 2014;592(14):2989–97. https://doi.org/10.1113/jphysiol.2014.273995.
136. Moloney RD, Desbonnet L, Clarke G, Dinan TG, Cryan JF. The microbiome: Stress, health and disease. Mamm Genome. 2014;25(1–2):49–74. https://doi.org/10.1007/s00335-013-9488-5.
137. Agudelo LZ, Femenía T, Orhan F, Porsmyr-Palmertz M, Goiny M, Martinez-Redondo V, et al. Skeletal muscle PGC-1α1 modulates kynurenine metabolism and mediates resilience to stress-induced depression. Cell. 2014;159(1):33–45. https://doi.org/10.1016/j.cell.2014.07.051.
138. Allison DJ, Nederveen JP, Snijders T, Bell KE, Kumbhare D, Phillips SM, et al. Exercise training impacts skeletal muscle gene expression related to the kynurenine pathway. Am J Physiol Cell Physiol. 2019;316(3):C444–8. https://doi.org/10.1152/ajpcell.00448.2018.
139. Bruce-Keller AJ, Salbaum JM, Luo M, Blanchard E, Taylor CM, Welsh DA, et al. Obese-type gut microbiota induce neurobehavioral changes in the absence of obesity. Biol Psychiatry. 2015;77(7):607–15. https://doi.org/10.1016/j.biopsych.2014.07.012.
140. Erickson MA, Dohi K, Banks WA. Neuroinflammation: a common pathway in CNS diseases as mediated at the blood-brain barrier. Neuroimmunomodulation. 2012;19(2):121–30. https://doi.org/10.1159/000330247.
141. Forsythe P, Sudo N, Dinan T, Taylor VH, Bienenstock J. Mood and gut feelings. Brain Behav Immun. 2010;24(1):9–16. https://doi.org/10.1016/j.bbi.2009.05.058.

142. Foster AC, Miller LP, Oldendorf WH, Schwarcz R. Studies on the disposition of quinolinic acid after intracerebral or systemic administration in the rat. Exp Neurol. 1984;84(2):428–40. https://doi.org/10.1016/0014-4886(84)90239-5.
143. Gourgouvelis J, Yielder P, Murphy B. Exercise promotes neuroplasticity in both healthy and depressed brains: an fMRI pilot study. Neural Plast. 2017;2017:1–13. https://doi.org/10.1155/2017/8305287.
144. Guillemin GJ. Quinolinic acid, the inescapable neurotoxin. FEBS J. 2012;279(8):1356–65. https://doi.org/10.1111/j.1742-4658.2012.08485.x.
145. Handschin C, Spiegelman BM. The role of exercise and PGC1α in inflammation and chronic disease. Nature. 2008;454(7203):463–9. https://doi.org/10.1038/nature07206.
146. Ignácio ZM, da Silva RS, Plissari ME, Quevedo J, Réus GZ. Physical exercise and neuroinflammation in major depressive disorder. Mol Neurobiol. 2019;56(12):8323–35. https://doi.org/10.1007/s12035-019-01670-1.
147. Javelle F, Bloch W, Knoop A, Guillemin GJ, Zimmer P. Toward a neuroprotective shift: Eight weeks of high intensity interval training reduces the neurotoxic kynurenine activity concurrently to impulsivity in emotionally impulsive humans – a randomized controlled trial. Brain Behav Immun. 2021;96:7–17. https://doi.org/10.1016/j.bbi.2021.04.020.
148. Kelly JR, Kennedy PJ, Cryan JF, Dinan TG, Clarke G, Hyland NP. Breaking down the barriers: the gut microbiome, intestinal permeability and stress-related psychiatric disorders. Front Cell Neurosci. 2015;9:392. https://doi.org/10.3389/fncel.2015.00392.
149. Kiliaan AJ, Saunders PR, Bijlsma PB, Cecilia Berin M, Taminiau JA, Groot JA, et al. Stress stimulates transepithelial macromolecular uptake in rat jejunum. Am J Physiol Gastrointest Liver Physiol. 1998;275:1037–44. https://doi.org/10.1152/ajpgi.1998.275.5.g1037.
150. Mota-Pereira J, Silverio J, Carvalho S, Ribeiro JC, Fonte D, Ramos J. Moderate exercise improves depression parameters in treatment-resistant patients with major depressive disorder. J Psychiatr Res. 2011;45(8):1005–11. https://doi.org/10.1016/j.jpsychires.2011.02.005.
151. Noble EE, Hsu TM, Kanoski SE. Gut to brain dysbiosis: mechanisms linking western diet consumption, the microbiome, and cognitive impairment. Front Behav Neurosci. 2017;11:9. https://doi.org/10.3389/fnbeh.2017.00009.
152. Obermeier B, Daneman R, Ransohoff RM. Development, maintenance and disruption of the blood-brain barrier. Nat Med. 2013;19(12):1584–96. https://doi.org/10.1038/nm.3407.
153. Phillips C, Fahimi A. Immune and neuroprotective effects of physical activity on the brain in depression. Front Neurosci. 2018;12:498. https://doi.org/10.3389/fnins.2018.00498.
154. Porras-Segovia A, Rivera M, Molina E, López-Chaves D, Gutiérrez B, Cervilla J. Physical exercise and body mass index as correlates of major depressive disorder in community-dwelling adults: results from the PISMA-ep study. J Affect Disord. 2019;251:263–9. https://doi.org/10.1016/j.jad.2019.01.050.
155. Roy Sarkar S, Banerjee S. Gut microbiota in neurodegenerative disorders. J Neuroimmunol. 2019 Mar;328:98–104. https://doi.org/10.1016/j.jneuroim.2019.01.004.
156. Ruas JL, White JP, Rao RR, Kleiner S, Brannan KT, Harrison BC, et al. A PGC-1α isoform induced by resistance training regulates skeletal muscle hypertrophy. Cell. 2012;151(6):1319–31. https://doi.org/10.1016/j.cell.2012.10.050.
157. Villena JA. New insights into PGC-1 coactivators: redefining their role in the regulation of mitochondrial function and beyond. FEBS J. 2015;282(4):647–72. https://doi.org/10.1111/febs.13175.

# 9 Glial-Neuronal Interaction in Synapses: A Possible Mechanism of the Pathophysiology of Bipolar Disorder

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**Abstract**

Bipolar disorder (BD) is a severe and chronic psychiatric disorder that affects approximately 1–4% of the world population and is characterized by recurrent episodes of mania or hypomania and depression. BD is also associated with illnesses marked by immune activation, such as metabolic syndrome, obesity,

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type 2 diabetes mellitus, and cardiovascular diseases. Indeed, a connection has been suggested between neuroinflammation and peripheral inflammatory markers in the pathophysiology of BD, which can be associated with the modulation of many dysfunctional processes, including synaptic plasticity, neurotransmission, neurogenesis, neuronal survival, apoptosis, and even cognitive/behavioral functioning. Rising evidence suggests that synaptic dysregulations, especially glutamatergic system dysfunction, are directly involved in mood disorders. It is becoming clear that dysregulations in connection and structural changes of glial cells play a central role in the BD pathophysiology. This book chapter highlighted the latest findings that support the theory of synaptic dysfunction in BD, providing an overview of the alterations in neurotransmitters release, astrocytic uptake, and receptor signaling, as well as the role of inflammation on glial cells in mood disorders. Particular emphasis is given to the alterations in presynaptic and postsynaptic neurons and glial cells, all cellular elements of the "tripartite synapse," compromising the neurotransmitters system, excitatory-inhibitory balance, and neurotrophic states of local networks in mood disorders. Together, these studies provide a foundation of knowledge about the exact role of the glial-neuronal interaction in mood disorders.

**Keywords**

Bipolar disorder · Glial cells · Astrocytes · Microglia · Neurons · Tripartite synapses

## 9.1 Introduction

Bipolar disorder (BD) is a severe and chronic psychiatric disorder that affects approximately 1–4% of the world population [1] and is characterized by recurrent episodes of mania or hypomania and depression [2]. BD is highly incapacitating and associated with premature mortality. Depressive symptoms and episodes are the most frequent cause of disability in patients with BD, and over half of patients do not respond adequately to approved treatments for this condition, showing the need for new classes of treatments to complement the current pharmacotherapy. In addition, long-term BD frequently leads to enduring functional and cognitive impairment [3, 4]. These changes occur both during the acute episodes of the disease, whether depressive or manic, and during euthymia, a state characterized by the remission of symptoms [5, 6]. While persistent cognitive deficits in BD are well established, there is significant heterogeneity in the literature regarding the specific domains of cognition that are affected, and not all patients seem to be equally affected. For example, 40% of BD patients have cognitive deficits in one to two domains, 22% are affected in three to five domains, and 38% of patients do not display any cognitive deficits [7]. Taken together, these studies indicate very heterogeneous cognitive deficits in some, but not all, BD patients, suggesting the possibility of subgroups existing among BD patients.

BD is also associated with illnesses marked by immune activation, such as metabolic syndrome [8], obesity, type 2 diabetes mellitus, and cardiovascular diseases [9], suggesting a connection between neuroinflammation and peripheral inflammatory markers in the pathophysiology of BD [10]. Indeed, evidence shows a significantly higher cytokine level in patients with BD [11], during acute mood episodes, euthymia, and after treatment [12]. Moreover, the chronic and mild inflammation observed in BD patients can trigger atherosclerosis, hypertension, and diabetes [13], comorbidities described in BD, suggesting that BD may be a multisystem disease with an inflammatory component, both peripherally and in the CNS. Several meta-analyses reported significantly higher concentrations of circulating pro-inflammatory cytokines such as interleukin 6 (IL-6) and tumor necrosis factor-α (TNF-α) in BD patients [11, 13, 14]. Moreover, studies have shown that immune system hyperactivation can be associated with modulating many dysfunctional processes in BD, including synaptic plasticity, neurotransmission, neurogenesis, neuronal survival, apoptosis, and even cognitive/behavioral functioning [15–18]. Immune system hyperactivation, propagated by increased serum interleukin-1β (IL-1β) and TNF-α levels, correlates with increased risk of depression and cognitive impairment and decreased treatment responsivity, especially to lithium. Additionally, peripheral sTNF-R1 levels positively correlate with disease severity, decreased cognitive function, and psychotic features in BD patients [16, 19].

Patel and colleagues [20] have suggested that the blood-brain-barrier (BBB) of patients with BD can be impaired, facilitating the passage of pro-inflammatory molecules from the periphery and decreasing central nervous system (CNS) protection. Furthermore, studies using neuroimaging and postmortem samples have found that BD patients present increased neuroinflammation through excessive microglial activation [20, 21]. Activation of glial cells dysregulates the innate immune system, such as the complement system, scavenger, and toll receptors, causing neuronal death [21]. In this context, this chapter aims to provide an overview of the recent findings in the field and provide a comprehensive update on the most recent hypotheses concerning the glial-neuronal interaction in BD.

## 9.2 BD and Glial Cells

Glial cells constitute between 33 and 66% of the total brain mass, depending on the mammalian species [22, 23]. In a very simplified view, glia cells provide support and protection for the neurons and can be subdivided into four major groups: microglia, astrocytes, oligodendrocytes, and their progenitors NG2-glia. Microglia play an essential role in remaining under constant surveillance, in a resting state, to protect the brain parenchyma (for review, see) [24–26]. It is known that microglia can exhibit two central activation phenotypes due to their high plasticity [27]. The classical activation is known as M1, which is the mediator of pro-inflammatory responses (hyper ramified or ameboid/phagocytic). The alternative activation, known as M2, is responsible for resolution and repair (anti-inflammatory). Thus,

the main functions of microglia in the adult brain are to monitor the environment and to start an inflammatory response in case of the detection of any dangerous signal. Microglia can also eliminate harmful debris and promote tissue repair and homeostasis, partly by affecting the surrounding astrocytes and neurons [28, 29]. Microglia can also release anti-inflammatory cytokines, enhance axonal regeneration and neurogenesis, and promote trophic support [30, 31].

Neural immune interactions and inflammatory processes are involved in the pathogenesis of psychiatric disorders [32]. In BD, the brain and the peripheral blood concentrations of pro-inflammatory cytokines were increased both in preclinical and clinical studies [14, 33]. It is believed that in mood disorders, there is a lack of balance between M1 and M2 phenotypes [26, 34]. A postmortem study indicates an increase in activated microglia density and macrophage recruitment in the cortex of depressed patients who committed suicide [35]. Neuroimaging studies reinforce that there is greater microglial activation in the prefrontal cortex, anterior cingulate, and insula in patients with BD during depressive episodes compared to control individuals [36]. The same evidence of neuroinflammation resulting from microglial activation has also been observed in the hippocampus of individuals with BD [37]. Pandey [38] found a significantly increased mRNA and protein expression of TNF-α, IL-1β, IL-6, and Toll-like receptors in postmortem prefrontal cortex tissues of suicide victims. Moreover, Pantazatos et al. [39] showed altered immune-related gene expression in depression and suicide and lower expression of genes associated with glial cell functions.

The microglia-mediated inflammatory processes can disrupt and damage glutamate homeostasis by astrocytic functions impairment [40, 41]. The action of these cytokines, after microglial activation, on the astrocyte occurs directly through disrupting the reuptake of the neurotransmitters by decreasing the expression of neurotransmitters transporters in astrocytes, increasing neurotransmitters release, tissue metabolism, and ROS production [42], which contributes to neuronal damage during neuroinflammation [43]. Therefore, it is believed that in BD, the immune system is chronically activated by microglia, which makes the CNS vulnerable and unstable to many varieties of insults, leading to mood disturbances [44]. Interestingly, treatment with antidepressants appears to modulate serum concentrations of inflammatory cytokines in individuals with mood disorders, resulting in lesser microglial activation [45]. Lower concentrations of IL-10 and chemokines are observed in individuals with major depressive disorder (MDD) who respond to antidepressant treatment, especially with selective serotonin reuptake inhibitors (SSRIs) [46, 47]. On the other hand, an increase in the concentration of inflammatory cytokines has been demonstrated in approximately one-third of patients who do not respond to treatment. In this sense, it is suggested that the lack of response to antidepressant therapy is due, in part, to a dysfunction of the immune system [40].

Inflammatory cytokines also play an essential role in activating the kynurenine pathway, which has been associated with BD. Briefly, enzymes in this pathway are located preferentially in glial cells [48]. Microglia express kynurenine 3-monooxygenase and produce quinolinic acid, while kynurenine aminotransferase is observed in astrocytes forming kynurenic acid. Kynurenic acid is a

neuroprotective molecule with nonselective antagonist activity at NMDA receptors and antioxidant properties [49]. On the other hand, quinolinic acid favors excitotoxicity by selectively activating NMDA receptors, favoring the release of glutamate while inhibiting the astrocytic reuptake of this neurotransmitter [49]. In BD, studies have found a reduction in the peripheral index of kynurenic acid/quinolinic acid, indicating a lower concentration of the neuroprotective compound [50, 51]. Furthermore, lower plasma concentrations of tryptophan and kynurenic acid observed in patients with mood disorders correlate with the severity of depressive symptoms and could function as a biomarker with predictive diagnostic potential [52]. In the same line, Benevenuto et al. [53] showed that kynurenine metabolites are decreased in both BD patients and unaffected BD offspring, and are associated with depression severity symptoms, suggesting that the kynurenine pathway might underlie the familial risk of BD shown by high-risk offspring individuals.

Astrocytes are the most abundant and highly distributed glial cells in the CNS, which have an intrinsic relationship with neurons as they are arranged in a network of interposed processes [54]. They act in the development and functions of the CNS, in synaptogenesis, in the ionic homeostasis of the extracellular environment and brain microcirculation, in the modulation of synaptic signaling, and also play an essential role in the metabolic support of neurons [55–57]. Glutamate homeostasis is dependent on two astrocytic functions: clearance of excess glutamate, preventing excitotoxicity, and glutamate storage and transportation to neurons in the form of glutamine provided by glial cells [54, 58]. Glial fibrillary acidic protein (GFAP) is a classical astrocyte protein marker [59, 60], and it is commonly altered in the brain of patients with BD [61], where a significant increase in GFAP expression in the dorsolateral prefrontal cortex of patients with BD was evidential [62]. GFAP mRNA levels have been shown to be increased in the peripheral blood of patients with BD not treated with lithium when compared with the lithium-treated BD patients and control subjects [63]. In an in vitro study, Vadodaria et al. [64] showed that BD hiPSCs-derived astrocytes generated from BD patients are transcriptionally distinct from controls and are less supportive for neuronal activity, even without stimulation. In the same study, the authors found that the addition of IL-6 blocking antibody in the conditioned culture medium of stimulated BD astrocytes was sufficient to rescue the decrease neuronal activity, suggesting that secreted factors from astrocytes play a role in regulating neuronal activity and that, in the case of BD, IL-6 at least in part mediated the effects of inflammation-primed astrocytes on neuronal activity. Moreover, a study showed that BD patients exhibit elevated levels of serum S100B during manic episodes [65]. The serum increment in S100B content represents an alteration in astrocyte activity, such as an alteration in GFAP expression. This alteration and other reported glial cells in mood disorders reinforce the involvement of astrocytes in the pathogenesis of BD [66–68]. However, like in other brain disorders, it remains to be shown whether the S100B increment reflects an astrocytic death or an active secretion of S100B to repair neuronal damage [69, 70].

## 9.3 BD and Neurons

Since the mid-twentieth century, the monoaminergic hypothesis, based on pharmacological studies, has been the most accepted and one of the primary explanations for the neurobiological basis of mood disorders [71]. Therefore, mood changes, especially depressive episodes, occur due to the reduction of central levels of monoamines serotonin (5-HT), dopamine (DA), and norepinephrine (NE) [72]. Supporting this theory, studies have shown that low concentrations of homovanillic acid (HVA) and 5-hydroxyindoleacetic acid (5-HIAA) in the CSF are associated with depressive symptoms in patients with mood disorders [73–75]. Young et al. [76] showed that although the levels of NE, 5-HT, and DA were not different in BD brain, an increase in the NE turnover and a decrease in 5-HT metabolites was found, suggesting an alteration in the balance of NE and 5-HT in mood disorders. Palsson et al. [77] showed that BD patients had significantly higher CSF levels of HVA and 5-HIAA but lower levels of 3-methoxy-4-hydroxyphenylglycol (MHPG) when compared to controls. The same authors also demonstrated that euthymic patients with a history of psychosis had an increased CSF concentration of HVA, while BD patients without such history did not differ significantly from controls [77, 78]. Furthermore, higher concentrations of MHPG in CSF have been associated with acute mania or atypical symptoms of depression in patients with BD [79, 80], while mood-stable patients had lower MHPG concentration [77]. Indeed, one of the main targets of current antidepressant therapies is the modulation of these neurotransmitters, which occurs mainly through the inhibition of reuptake or inhibition of the metabolism of monoamines [72].

However, the monoaminergic hypothesis can no longer fully elucidate the pathophysiology of mood disorders. The balance between excitatory and inhibitory impulses is assumed to be essential for processing information and preserving cognitive functions. Therefore, new therapies have been developed to modulate the neurotransmission of gamma-aminobutyric acid (GABA) and glutamate, the primary inhibitory and excitatory neurotransmitters, respectively [81]. The dysregulation in the levels of these primary impulses leads to a change in neural activity and resting state, which chronically can result in a maladaptation of these neuronal systems, contributing to the onset of mood symptoms [82]. The exact ratio between GABA and glutamate seems to be altered in patients with BD [83]. Unfortunately, there is still no consensus on both peripheral and central levels of these neurotransmitters in patients with mood disorders due to the difficulty of studies in controlling the effects of medication and postmortem metabolism [84]. Even so, patients who show a reduction in cortical GABA seem to have a more significant cognitive impairment, especially concerning inhibitory control [85, 86]. On the other hand, high levels of glutamate have already been observed in the prefrontal cortex of individuals with BD [87, 88]. Furthermore, a neuropathological study showed a significant decrease in the density of glutamate receptor ASCT-1 (neutral amino acid transporter 1) in neurons and glial cells in BD patients compared to healthy controls [89].

Glutamate receptor modulators, such as ketamine, have been suggested as potential treatments for BD depression because of their rapid-acting and sustained antidepressant effects [90, 91]. The first study of ketamine in BD depression showed the effect of ketamine as an adjunct to mood stabilizers (lithium or valproate) in patients. A difference was observed within 40 min after infusion, and this improvement remained significant through day 3 [92]. Replication of these results occurred in 2012 after infusion of ketamine demonstrated a clinical response of 79% effectiveness [93], and on the 7th day after a single infusion of ketamine in half of the patients with bipolar depression receiving mood stabilizers, in which treatment with antidepressants had not had a satisfactory effect [94]. It is speculated that a single ketamine infusion could improve neuropsychological performance independently of its antidepressant effect [95]. Recently, a meta-analysis found higher antidepressant response rates in patients with BD and improvement in suicidal ideation after ketamine administration [96]. Ketamine acts on several pharmacologic targets, having effects on glutamatergic transmission, BDNF levels, and intracellular signal transduction, which are all perturbed in patients with BD. Moreover, ketamine also shows beneficial effects on synaptogenesis and neuroplasticity, and the ability to regulate inflammation [97].

## 9.4 Neuron-Glia Interactions in BD

The bidirectional communication between neurons and astrocytes on synapses is called "tripartite synapse," which comprises three cellular elements: postsynaptic and presynaptic neurons and astrocytes [98]. There is a common agreement regarding the fact that astrocytes are considered morphological and metabolic support cells since they play a crucial role in the synthesis and reuptake of glutamate, in buffering extracellular $K^+$ to control neuronal excitability, in the release of gliotransmitters, facilitating neuro-energetics, participating in cerebral inflammation, and in the maintenance of the BBB [99, 100]. Moreover, astrocytes display dynamic signaling with neurons and synapses by sensing the neuronal and synaptic activity through ion channel activation and neurotransmitter transporters and receptors. Recently, microglia have been added to the number of synaptic players, creating the "quad-partite" synapse. As described above, microglia coordinate brain innate immunity, displaying features characteristic of immune cells able to rapidly expand their population, migrate to injury sites, and trigger and sustain inflammatory responses through their chemokine and cytokine repertoire [101, 102]. Microglia interact with neurons and astrocytes in the resting state to support and regulate brain homeostasis, acting in excitatory and inhibitory transmission by releasing chemokines, cytokines, purines, glutamate D-serine, ATP, and BDNF [103–108].

Growing evidence points to the hypothesis that all elements of the quad-partite glutamatergic synapse may be altered in BD. Briefly, in glutamatergic synapses, following presynaptic neuronal depolarization, calcium channels open, permitting the influx of calcium and triggering synaptic vesicles loaded with glutamate, by the vesicular glutamate transporter (vGluT) to fuse with the presynaptic membrane by

interacting with soluble N-ethylmaleimide-sensitive factor attachment protein receptors (SNARE), creating a large opening through which glutamate is released into the synaptic cleft [54, 109]. Upon release, glutamate binds to and activates ionotropic and metabotropic receptors, resulting in both immediate changes in membrane potential and sustained alterations in synaptic connectivity. Because there are no extracellular enzymes to degrade glutamate, the only way to terminate glutamate signaling, and to keep extracellular glutamate levels low, is through uptake of glutamate by excitatory amino acid transporters (EAAT), especially EAAT2, encoded by the SLC1A2 gene, on neighboring astrocytes [99, 110]. In glial cells, glutamate is converted to glutamine catalyzed by glutamine synthetase, which is later returned to neurons to generate glutamate, completing the glutamate-glutamine cycle (Fig. 9.1). Thus, EAAT plays a crucial role in preventing extracellular glutamate concentrations from reaching neurotoxic levels and recycling glutamate at synapses by transporting glutamate into astrocytes for conversion to glutamine [111]. In BD, studies have shown that the SLC1A2 promoter region was hypermethylated in BD patients [112] and that the SLC1A2 polymorphisms (rs4354668) in BD patients with low scores of adverse childhood experiences significantly influence gray matter [113]. Besides, magnetic resonance spectroscopy (MRS) studies have shown that patients with BD had elevated brain glutamate/glutamine ratio [114–118].

Numerous molecular factors indicate region-specific alterations of presynaptic functions in BD. Eastwood and Harrison [119] found that vGluT1, netrin-G2, netrin-G1d, and netrin-G1f mRNA levels were elevated in the anterior cingulate cortex in patients with BD. On the other hand, a decrease in the vGluT1 and netrin-G1c mRNA expression was found in the entorhinal and temporal cortex of patients with BD [120, 121]. Postmortem studies also indicate abnormalities in the expression of individual SNARE proteins and regulatory proteins in the hippocampus and frontal cortex of patients with depression and BD [122, 123]. Moreover, studies have also found associations between mood stabilizers and presynaptic markers. Kim and Thayer [124] showed that lithium-induced inositol depletion increases the formation of new synapses between hippocampal neurons and an increase in fluorescent puncta formed by the presynaptic marker synaptophysin-GFP. The same authors also showed that the inhibition of postsynaptic NMDA receptors or presynaptic calcium channels significantly reduced lithium-induced synapse formation, indicating that glutamatergic synaptic transmission was required for the effects of lithium [124]. More recently, a study showed that chronic lithium treatment significantly reduced intracellular calcium flux in mouse cortical neurons by activating mGluR5 [125]. The same study demonstrated that chronic lithium reduced spine number and decreased the percentage of mature spines, mature spine width, and PSD-95 puncta intensity. Ketamine also inhibits glutamate transmission from astrocytes to neurons and disrupts the synchronization of astrocytic slow inward currents, presumably mediated by the extrasynaptic GluN1/GluN2B receptors [126]. Evidence from preclinical studies also demonstrated that ketamine rapidly induces changes in the hippocampal presynaptic machinery, including a downregulation in CaMKIIa phosphorylation, which consequently reduced its binding to syntaxin 1A, therefore,

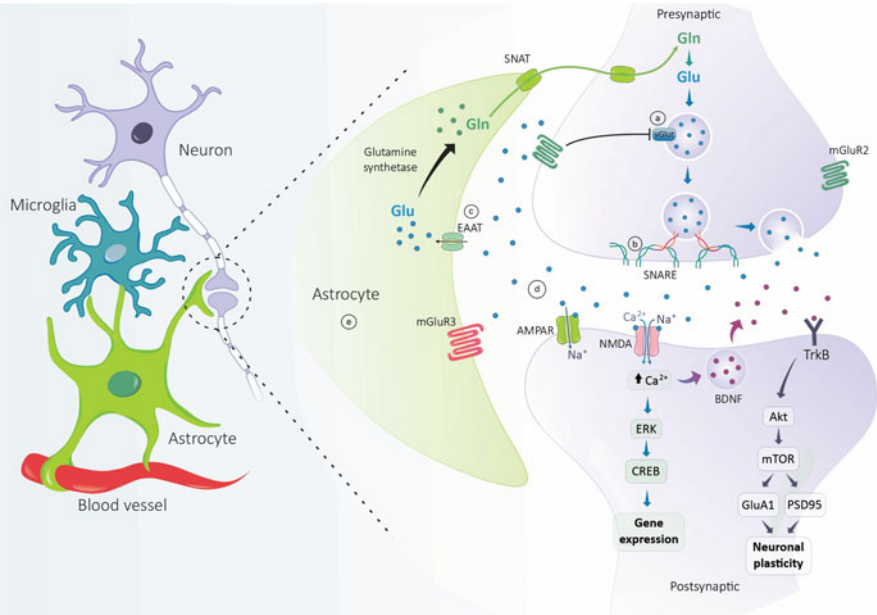


**Fig. 9.1** Schematic representation of tripartite glutamatergic synapses and alterations observed in BD. Glutamate (Glu) is synthesized in presynaptic neurons and is loaded into synaptic vesicles via the vesicular glutamate transporter (VGluT). SNARE complex proteins mediate the fusion of vesicles with the presynaptic membrane. After release into the synaptic cleft, glutamate binds to ionotropic glutamate receptors (NMDA re and AMPAR) and metabotropic glutamate receptors (mGluR1 to mGluR8) on both postsynaptic and presynaptic neurons, resulting in both immediate changes, including membrane depolarization, activation of intracellular messenger cascades, modulation of local protein synthesis, and, eventually, gene expression. Glutamate in the extracellular space is taken up by astrocytes through (EAAT1 and EAAT2), whose expression in astrocytes can be upregulated by glutamate. The conversion of glutamate into glutamine is catalyzed by glutamine synthetase and glutamine is transported back to the presynaptic neuron to be converted back to glutamate. Studies have associated several changes in all elements of the tripartite glutamatergic synapse in BD, not all of which are shown here. (**a**) Changes in the expression of vGluT (**b**) abnormalities in the expression of SNARE proteins; (**c**) reduced glutamate clearance by EAATs; (**d**) glutamate spillover, which leads to increased activation of extrasynaptic glutamate receptors, resulting in excitotoxicity; (**e**) increase in the expression of the glial fibrillary acid protein (GFAP)

interfering with SNARE complex assembly, as well as a decrease in the expression of the synaptic vesicle protein synaptotagmin I and an increase in the levels of synapsin I in hippocampal synaptosomes [127, 128].

Additionally, evidence for glial involvement in BD has also been described. However, findings concerning glial cells' overall number and density in specific subregions are contradictory [68, 129–133]. A recent meta-analysis showed findings of glial deficits and a thinning of grey matter, as well as a reduced density of CB-positive neurons in some layers of the dorsolateral prefrontal cortex (DLPFC), suggesting that interneurons may be affected in BD. Studies have also described reductions in neuronal markers in several brain regions [134, 135]. Tobe et al. [136]

showed that dendrite length and spine density is diminished in postmortem brain tissue from the DLPFC of patients with BD, but no changes in spine density were observed in individuals BD patients treated with lithium. Furthermore, reductions of parvalbumin- and somatostatin-positive interneurons in the parahippocampal area, lateral amygdala nucleus, and thalamic reticular nucleus in BD have been described, which could be associated with disrupting synchronization and integration of cortico-hippocampal circuits [137–139]. Using in vitro models by differentiating mature neurons from human-induced pluripotent stem cells (hiPSCs) derived from BD patients with a PCDH15 (protocadherin related 15) deletion, a study found that hi-PSCs-derived glutamatergic neurons exhibited abnormalities in dendrite and synapse formation [140]. Using a similar in vitro model, Kim et al. [141] showed that the expression of GAD1, the gene encoding glutamate acid decarboxylase, in hiPSCs-derived neurons from BD patients was increased.

In view of the crucial role of aberrant synaptic plasticity involving neuronal, astrocytic, and microglia dysfunction, Mitterauer [142] proposed a model of imbalances in tripartite synapses in mood disorders based on a formalism of system-balancing. Thus, the expression of astroglial receptors would determine the imbalances of neurotransmission. Based on this model, in depression, the upregulation of gap junctions exerts an overexpression of astroglial receptors that cannot be activated by neurotransmitters, which leads to low $Ca^{2+}$ levels and underproduction of gliotransmitters resulting in prolonged neurotransmission. On the other hand, in mania, the imbalance of tripartite synapses is caused by an under-expression of gap junctions in the astroglial network and astroglial receptors, which causes an increase in $Ca^{2+}$ levels and gliotransmitters that exert shortened feedback on the presynaptic receptors causing a shortened neurotransmission. However, this model is mainly theoretical and must be clinically and biologically tested.

## 9.5 Conclusion

This compilation of evidence illustrates the relevance of synaptic dysfunction, in particular, the role played by the tripartite or quad-partite glutamatergic synapse (presynaptic neuron, postsynaptic neuron, and glia) in the pathophysiology of mood disorders. However, the mechanisms leading to tripartite synapse dysregulation remain to be determined. Thus, an increase in understanding of the role of neuron-glia interaction in mood disorders is highly warranted. Future studies need to look at more developmental time points, different brain regions, and synapse types and consider the astrocyte heterogeneity. Improving our understanding of these alterations may provide the framework to investigate the complex mechanisms behind mood disorders and novel therapeutic options.

## References


1. Grande I, Berk M, Birmaher B, Vieta E. Bipolar disorder. Lancet. 2016;387(10027):1561–72.
2. Severus E, Bauer M. Bipolar disorders in DSM-5. Nervenarzt. 2014;85(5):543–7.
3. Panchal P, Kaltenboeck A, Harmer CJ. Cognitive emotional processing across mood disorders. CNS Spectr. 2019;24(1):54–63.
4. Passos IC, Mwangi B, Vieta E, Berk M, Kapczinski F. Areas of controversy in neuroprogression in bipolar disorder. Acta Psychiatr Scand. 2016;134(2):91–103.
5. Martinez-Aran A, Vieta E, Torrent C, Sanchez-Moreno J, Goikolea JM, Salamero M, et al. Functional outcome in bipolar disorder: the role of clinical and cognitive factors. Bipolar Disord. 2007;9(1-2):103–13.
6. Petersen JZ, Porter RJ, Miskowiak KW. Clinical characteristics associated with the discrepancy between subjective and objective cognitive impairment in depression. J Affect Disord. 2019;246:763–74.
7. Martino DJ, Strejilevich SA, Scapola M, Igoa A, Marengo E, Ais ED, et al. Heterogeneity in cognitive functioning among patients with bipolar disorder. J Affect Disord. 2008;109(1-2):149–56.
8. Godin O, Etain B, Henry C, Bougerol T, Courtet P, Mayliss L, et al. Metabolic syndrome in a French cohort of patients with bipolar disorder: results from the FACE-BD cohort. J Clin Psychiatry. 2014;75(10):1078–85; quiz 85.
9. Jerrell JM, McIntyre RS, Tripathi A. A cohort study of the prevalence and impact of comorbid medical conditions in pediatric bipolar disorder. J Clin Psychiatry. 2010;71(11):1518–25.
10. Berk M, Kapczinski F, Andreazza AC, Dean OM, Giorlando F, Maes M, et al. Pathways underlying neuroprogression in bipolar disorder: focus on inflammation, oxidative stress and neurotrophic factors. Neurosci Biobehav Rev. 2011;35(3):804–17.
11. Munkholm K, Brauner JV, Kessing LV, Vinberg M. Cytokines in bipolar disorder vs. healthy control subjects: a systematic review and meta-analysis. J Psychiatr Res. 2013;47(9):1119–33.
12. Goldsmith DR, Rapaport MH, Miller BJ. A meta-analysis of blood cytokine network alterations in psychiatric patients: comparisons between schizophrenia, bipolar disorder and depression. Mol Psychiatry. 2016;21(12):1696–709.
13. Goldstein BI, Kemp DE, Soczynska JK, McIntyre RS. Inflammation and the phenomenology, pathophysiology, comorbidity, and treatment of bipolar disorder: a systematic review of the literature. J Clin Psychiatry. 2009;70(8):1078–90.
14. Modabbernia A, Taslimi S, Brietzke E, Ashrafi M. Cytokine alterations in bipolar disorder: a meta-analysis of 30 studies. Biol Psychiatry. 2013;74(1):15–25.
15. Guloksuz S, Altinbas K, Aktas Cetin E, Kenis G, Bilgic Gazioglu S, Deniz G, et al. Evidence for an association between tumor necrosis factor-alpha levels and lithium response. J Affect Disord. 2012;143(1-3):148–52.
16. Hoseth EZ, Ueland T, Dieset I, Birnbaum R, Shin JH, Kleinman JE, et al. A study of TNF pathway activation in schizophrenia and bipolar disorder in plasma and brain tissue. Schizophr Bull. 2017;43(4):881–90.
17. Khairova RA, Machado-Vieira R, Du J, Manji HK. A potential role for pro-inflammatory cytokines in regulating synaptic plasticity in major depressive disorder. Int J Neuropsychopharmacol. 2009;12(4):561–78.
18. Panaccione I, Spalletta G, Sani G. Neuroinflammation and excitatory symptoms in bipolar disorder. Neuroimmunol Neuroinflamm. 2015;2:215–27.
19. Hope S, Ueland T, Steen NE, Dieset I, Lorentzen S, Berg AO, et al. Interleukin 1 receptor antagonist and soluble tumor necrosis factor receptor 1 are associated with general severity and psychotic symptoms in schizophrenia and bipolar disorder. Schizophr Res. 2013;145(1-3):36–42.
20. Patel JP, Frey BN. Disruption in the blood-brain barrier: the missing link between brain and body inflammation in bipolar disorder? Neural Plast. 2015;2015:708306.

21. Hauwel M, Furon E, Canova C, Griffiths M, Neal J, Gasque P. Innate (inherent) control of brain infection, brain inflammation and brain repair: the role of microglia, astrocytes, "protective" glial stem cells and stromal ependymal cells. Brain Res Brain Res Rev. 2005;48(2): 220–33.
22. Azevedo FA, Carvalho LR, Grinberg LT, Farfel JM, Ferretti RE, Leite RE, et al. Equal numbers of neuronal and nonneuronal cells make the human brain an isometrically scaled-up primate brain. J Comp Neurol. 2009;513(5):532–41.
23. Herculano-Houzel S. The glia/neuron ratio: how it varies uniformly across brain structures and species and what that means for brain physiology and evolution. Glia. 2014;62(9):1377–91.
24. Boche D, Perry VH, Nicoll JA. Review: activation patterns of microglia and their identification in the human brain. Neuropathol Appl Neurobiol. 2013;39(1):3–18.
25. Brisch R, Wojtylak S, Saniotis A, Steiner J, Gos T, Kumaratilake J, et al. The role of microglia in neuropsychiatric disorders and suicide. Eur Arch Psychiatry Clin Neurosci. 2021;272(6): 929–45.
26. Nakagawa Y, Chiba K. Role of microglial m1/m2 polarization in relapse and remission of psychiatric disorders and diseases. Pharmaceuticals. 2014;7(12):1028–48.
27. Franco R, Fernandez-Suarez D. Alternatively activated microglia and macrophages in the central nervous system. Prog Neurobiol. 2015;131:65–86.
28. Herzog C, Pons Garcia L, Keatinge M, Greenald D, Moritz C, Peri F, et al. Rapid clearance of cellular debris by microglia limits secondary neuronal cell death after brain injury in vivo. Development. 2019;146(9):174698.
29. Neumann H, Kotter MR, Franklin RJ. Debris clearance by microglia: an essential link between degeneration and regeneration. Brain. 2009;132:288–95.
30. Gomes-Leal W. Microglial physiopathology: how to explain the dual role of microglia after acute neural disorders? Brain Behav. 2012;2(3):345–56.
31. Kalafatakis I, Karagogeos D. Oligodendrocytes and microglia: key players in myelin development, damage and repair. Biomol Ther. 2021;11(7):1058.
32. Berk M, Williams LJ, Jacka FN, O'Neil A, Pasco JA, Moylan S, et al. So depression is an inflammatory disease, but where does the inflammation come from? BMC Med. 2013;11:200.
33. Wang AK, Miller BJ. Meta-analysis of cerebrospinal fluid cytokine and tryptophan catabolite alterations in psychiatric patients: comparisons between schizophrenia, bipolar disorder, and depression. Schizophr Bull. 2018;44(1):75–83.
34. Rahimian R, Wakid M, O'Leary LA, Mechawar N. The emerging tale of microglia in psychiatric disorders. Neurosci Biobehav Rev. 2021;131:1–29.
35. Torres-Sanchez S, Perez-Caballero L, Berrocoso E. Cellular and molecular mechanisms triggered by deep brain stimulation in depression: a preclinical and clinical approach. Prog Neuro-psychopharmacol Biol Psychiatry. 2017;73:1–10.
36. Qiu L, Ye J, Ji F, Li G, Li G, Ma X, et al. Common and distinct global functional connectivity density alterations in patients with bipolar disorder with and without auditory verbal hallucination during major depressive episodes. Brain Imaging Behav. 2020;14(6):2724–30.
37. Haarman BC, Burger H, Doorduin J, Renken RJ, Sibeijn-Kuiper AJ, Marsman JB, et al. Volume, metabolites and neuroinflammation of the hippocampus in bipolar disorder - a combined magnetic resonance imaging and positron emission tomography study. Brain Behav Immun. 2016;56:21–33.
38. Pandey GN. Inflammatory and innate immune markers of neuroprogression in depressed and teenage suicide brain. Mod Trends Pharmacopsychiatry. 2017;31:79–95.
39. Pantazatos SP, Huang YY, Rosoklija GB, Dwork AJ, Arango V, Mann JJ. Whole-transcriptome brain expression and exon-usage profiling in major depression and suicide: evidence for altered glial, endothelial and ATPase activity. Mol Psychiatry. 2017;22(5): 760–73.
40. Haroon E, Daguanno AW, Woolwine BJ, Goldsmith DR, Baer WM, Wommack EC, et al. Antidepressant treatment resistance is associated with increased inflammatory markers in patients with major depressive disorder. Psychoneuroendocrinology. 2018;95:43–9.

41. Mechawar N, Savitz J. Neuropathology of mood disorders: do we see the stigmata of inflammation? Transl Psychiatry. 2016;6(11):e946.
42. Efremova L, Chovancova P, Adam M, Gutbier S, Schildknecht S, Leist M. Switching from astrocytic neuroprotection to neurodegeneration by cytokine stimulation. Arch Toxicol. 2017;91(1):231–46.
43. Barger SW, Goodwin ME, Porter MM, Beggs ML. Glutamate release from activated microglia requires the oxidative burst and lipid peroxidation. J Neurochem. 2007;101(5):1205–13.
44. Schroeter ML, Abdul-Khaliq H, Sacher J, Steiner J, Blasig IE, Mueller K. Mood disorders are glial disorders: evidence from in vivo studies. Cardiovasc Psychiatry Neurol. 2010;2010: 780645.
45. Anderson G, Maes M. Bipolar disorder: role of immune-inflammatory cytokines, oxidative and nitrosative stress and tryptophan catabolites. Curr Psychiatry Rep. 2015;17(2):8.
46. Buspavanich P, Adli M, Himmerich H, Berger M, Busche M, Schlattmann P, et al. Faster speed of onset of the depressive episode is associated with lower cytokine serum levels (IL-2, -4, -6, -10, TNF-alpha and IFN-gamma) in patients with major depression. J Psychiatr Res. 2021;141:287–92.
47. Lindqvist D, Dhabhar FS, James SJ, Hough CM, Jain FA, Bersani FS, et al. Oxidative stress, inflammation and treatment response in major depression. Psychoneuroendocrinology. 2017;76:197–205.
48. Maddison DC, Giorgini F. The kynurenine pathway and neurodegenerative disease. Semin Cell Dev Biol. 2015;40:134–41.
49. Schwarcz R, Bruno JP, Muchowski PJ, Wu HQ. Kynurenines in the mammalian brain: when physiology meets pathology. Nat Rev Neurosci. 2012;13(7):465–77.
50. Ramirez LA, Perez-Padilla EA, Garcia-Oscos F, Salgado H, Atzori M, Pineda JC. A new theory of depression based on the serotonin/kynurenine relationship and the hypothalamicpituitary-adrenal axis. Biomedica. 2018;38(3):437–50.
51. Savitz J, Dantzer R, Wurfel BE, Victor TA, Ford BN, Bodurka J, et al. Neuroprotective kynurenine metabolite indices are abnormally reduced and positively associated with hippocampal and amygdalar volume in bipolar disorder. Psychoneuroendocrinology. 2015;52:200–11.
52. Liu H, Ding L, Zhang H, Mellor D, Wu H, Zhao D, et al. The metabolic factor kynurenic acid of kynurenine pathway predicts major depressive disorder. Front Psych. 2018;9:552.
53. Benevenuto D, Saxena K, Fries GR, Valvassori SS, Kahlon R, Saxena J, et al. Alterations in plasma kynurenine pathway metabolites in children and adolescents with bipolar disorder and unaffected offspring of bipolar parents: a preliminary study. Bipolar Disord. 2021;23(7): 689–96.
54. Mahmoud S, Gharagozloo M, Simard C, Gris D. Astrocytes maintain glutamate homeostasis in the CNS by controlling the balance between glutamate uptake and release. Cell. 2019;8(2): 184.
55. Giovannoni F, Quintana FJ. The role of astrocytes in CNS inflammation. Trends Immunol. 2020;41(9):805–19.
56. Pellerin L, Magistretti PJ. Glutamate uptake into astrocytes stimulates aerobic glycolysis: a mechanism coupling neuronal activity to glucose utilization. Proc Natl Acad Sci U S A. 1994;91(22):10625–9.
57. Pellerin L, Pellegri G, Bittar PG, Charnay Y, Bouras C, Martin JL, et al. Evidence supporting the existence of an activity-dependent astrocyte-neuron lactate shuttle. Dev Neurosci. 1998;20 (4-5):291–9.
58. Diaz-Ruiz A, Salgado-Ceballos H, Montes S, Maldonado V, Tristan L, Alcaraz-Zubeldia M, et al. Acute alterations of glutamate, glutamine, GABA, and other amino acids after spinal cord contusion in rats. Neurochem Res. 2007;32(1):57–63.
59. Colombo E, Farina C. Astrocytes: key regulators of neuroinflammation. Trends Immunol. 2016;37(9):608–20.

60. Yang Z, Wang KK. Glial fibrillary acidic protein: from intermediate filament assembly and gliosis to neurobiomarker. Trends Neurosci. 2015;38(6):364–74.
61. Rao JS, Harry GJ, Rapoport SI, Kim HW. Increased excitotoxicity and neuroinflammatory markers in postmortem frontal cortex from bipolar disorder patients. Mol Psychiatry. 2010;15 (4):384–92.
62. Feresten AH, Barakauskas V, Ypsilanti A, Barr AM, Beasley CL. Increased expression of glial fibrillary acidic protein in prefrontal cortex in psychotic illness. Schizophr Res. 2013;150(1): 252–7.
63. Ferensztajn-Rochowiak E, Tarnowski M, Samochowiec J, Michalak M, Ratajczak MZ, Rybakowski JK. Increased mRNA expression of peripheral glial cell markers in bipolar disorder: the effect of long-term lithium treatment. Eur Neuropsychopharmacol. 2016;26(9): 1516–21.
64. Vadodaria KC, Mendes APD, Mei A, Racha V, Erikson G, Shokhirev MN, et al. Altered neuronal support and inflammatory response in bipolar disorder patient-derived astrocytes. Stem Cell Rep. 2021;16(4):825–35.
65. Machado-Vieira R, Lara DR, Portela LV, Goncalves CA, Soares JC, Kapczinski F, et al. Elevated serum S100B protein in drug-free bipolar patients during first manic episode: a pilot study. Eur Neuropsychopharmacol. 2002;12(3):269–72.
66. Dong XH, Zhen XC. Glial pathology in bipolar disorder: potential therapeutic implications. CNS Neurosci Therap. 2015;21(5):393–7.
67. Keshavarz M. Glial cells as key elements in the pathophysiology and treatment of bipolar disorder. Acta Neuropsychiatrica. 2017;29(3):140–52.
68. Ongur D, Drevets WC, Price JL. Glial reduction in the subgenual prefrontal cortex in mood disorders. Proc Natl Acad Sci U S A. 1998;95(22):13290–5.
69. Hetzel G, Moeller O, Evers S, Erfurth A, Ponath G, Arolt V, et al. The astroglial protein S100B and visually evoked event-related potentials before and after antidepressant treatment. Psychopharmacology. 2005;178(2-3):161–6.
70. Van Eldik LJ, Wainwright MS. The Janus face of glial-derived S100B: beneficial and detrimental functions in the brain. Restor Neurol Neurosci. 2003;21(3-4):97–108.
71. Liu Y, Zhao J, Guo W. Emotional roles of mono-aminergic neurotransmitters in major depressive disorder and anxiety disorders. Front Psychol. 2018;9:2201.
72. Heninger GR, Delgado PL, Charney DS. The revised monoamine theory of depression: a modulatory role for monoamines, based on new findings from monoamine depletion experiments in humans. Pharmacopsychiatry. 1996;29(1):2–11.
73. Kasa K, Otsuki S, Yamamoto M, Sato M, Kuroda H, Ogawa N. Cerebrospinal fluid gamma-aminobutyric acid and homovanillic acid in depressive disorders. Biol Psychiatry. 1982;17(8): 877–83.
74. Peabody CA, Faull KF, King RJ, Whiteford HA, Barchas JD, Berger PA. CSF amine metabolites and depression. Psychiatry Res. 1987;21(1):1–7.
75. Reddy PL, Khanna S, Subhash MN, Channabasavanna SM, Rao BS. CSF amine metabolites in depression. Biol Psychiatry. 1992;31(2):112–8.
76. Young LT, Warsh JJ, Kish SJ, Shannak K, Hornykeiwicz O. Reduced brain 5-HT and elevated NE turnover and metabolites in bipolar affective disorder. Biol Psychiatry. 1994;35(2):121–7.
77. Palsson E, Sellgren C, Ryden E, Kizza R, Pelanis A, Zetterberg H, et al. Cerebrospinal fluid monoamine metabolite profiles in bipolar disorder, ADHD, and controls. J Neural Transm. 2017;124(9):1135–43.
78. Sellgren CM, Kegel ME, Bergen SE, Ekman CJ, Olsson S, Larsson M, et al. A genome-wide association study of kynurenic acid in cerebrospinal fluid: implications for psychosis and cognitive impairment in bipolar disorder. Mol Psychiatry. 2016;21(10):1342–50.
79. Redmond DE, Katz MM, Maas JW, Swann A, Casper R, Davis JM. Cerebrospinal fluid amine metabolites. Relationships with behavioral measurements in depressed, manic, and healthy control subjects. Arch Gen Psychiatry. 1986;43(10):938–47.

80. Swann AC, Secunda S, Davis JM, Robins E, Hanin I, Koslow SH, et al. CSF monoamine metabolites in mania. Am J Psychiatry. 1983;140(4):396–400.
81. Wilkinson ST, Sanacora G. A new generation of antidepressants: an update on the pharmaceutical pipeline for novel and rapid-acting therapeutics in mood disorders based on glutamate/GABA neurotransmitter systems. Drug Discov Today. 2019;24(2):606–15.
82. Fee C, Banasr M, Sibille E. Somatostatin-positive gamma-aminobutyric acid interneuron deficits in depression: cortical microcircuit and therapeutic perspectives. Biol Psychiatry. 2017;82(8):549–59.
83. Scotti-Muzzi E, Chile T, Moreno R, Pastorello BF, da Costa LC, Henning A, et al. ACC Glu/GABA ratio is decreased in euthymic bipolar disorder I patients: possible in vivo neurometabolite explanation for mood stabilization. Eur Arch Psychiatry Clin Neurosci. 2021;271(3):537–47.
84. Brady RO Jr, McCarthy JM, Prescot AP, Jensen JE, Cooper AJ, Cohen BM, et al. Brain gamma-aminobutyric acid (GABA) abnormalities in bipolar disorder. Bipolar Disord. 2013;15(4):434–9.
85. Duman RS, Sanacora G, Krystal JH. Altered connectivity in depression: GABA and glutamate neurotransmitter deficits and reversal by novel treatments. Neuron. 2019;102(1):75–90.
86. Luscher B, Shen Q, Sahir N. The GABAergic deficit hypothesis of major depressive disorder. Mol Psychiatry. 2011;16(4):383–406.
87. Nery FG, Tallman MJ, Cecil KM, Blom TJ, Patino LR, Adler CM, et al. N-acetylcysteine for depression and glutamate changes in the left prefrontal cortex in adolescents and young adults at risk for bipolar disorder: a pilot study. Early Interv Psychiatry. 2022;16(2):195–9.
88. Smaragdi A, Chavez S, Lobaugh NJ, Meyer JH, Kolla NJ. Differential levels of prefrontal cortex glutamate+glutamine in adults with antisocial personality disorder and bipolar disorder: a proton magnetic resonance spectroscopy study. Prog Neuro-psychopharmacol Biol Psychiatry. 2019;93:250–5.
89. Weis S, Llenos IC, Dulay JR, Verma N, Sabunciyan S, Yolken RH. Changes in region- and cell type-specific expression patterns of neutral amino acid transporter 1 (ASCT-1) in the anterior cingulate cortex and hippocampus in schizophrenia, bipolar disorder and major depression. J Neural Transm. 2007;114(2):261–71.
90. Berman RM, Cappiello A, Anand A, Oren DA, Heninger GR, Charney DS, et al. Antidepressant effects of ketamine in depressed patients. Biol Psychiatry. 2000;47(4):351–4.
91. Vande Voort JL, Ballard ED, Luckenbaugh DA, Bernert RA, Richards EM, Niciu MJ, et al. Antisuicidal response following ketamine infusion is associated with decreased nighttime wakefulness in major depressive disorder and bipolar disorder. J Clin Psychiatry. 2017;78(8):1068–74.
92. Diazgranados N, Ibrahim L, Brutsche NE, Newberg A, Kronstein P, Khalife S, et al. A randomized add-on trial of an N-methyl-D-aspartate antagonist in treatment-resistant bipolar depression. Arch Gen Psychiatry. 2010;67(8):793–802.
93. Zarate CA, Brutsche NE, Ibrahim L, Franco-Chaves J, Diazgranados N, Cravchik A, et al. Replication of ketamine's antidepressant efficacy in bipolar depression: a randomized controlled add-on trial. Biol Psychiatry. 2012;71(11):939–46.
94. Rybakowski JK, Permoda-Osip A, Bartkowska-Sniatkowska A. Ketamine augmentation rapidly improves depression scores in inpatients with treatment-resistant bipolar depression. Int J Psychiatry Clin Pract. 2017;21(2):99–103.
95. Permoda-Osip A, Kisielewski J, Bartkowska-Sniatkowska A, Rybakowski JK. Single ketamine infusion and neurocognitive performance in bipolar depression. Pharmacopsychiatry. 2015;48(2):78–9.
96. Bahji A, Pierce M, Wong J, Roberge JN, Ortega I, Patten S. Comparative efficacy and acceptability of psychotherapies for self-harm and suicidal behavior among children and adolescents: a systematic review and network meta-analysis. JAMA Netw Open. 2021;4(4):e216614.

97. Wilkowska A, Szalach L, Cubala WJ. Ketamine in bipolar disorder: a review. Neuropsychiatr Dis Treat. 2020;16:2707–17.
98. Araque A, Parpura V, Sanzgiri RP, Haydon PG. Tripartite synapses: glia, the unacknowledged partner. Trends Neurosci. 1999;22(5):208–15.
99. Farhy-Tselnicker I, Allen NJ. Astrocytes, neurons, synapses: a tripartite view on cortical circuit development. Neural Dev. 2018;13(1):7.
100. Haydon PG, Carmignoto G. Astrocyte control of synaptic transmission and neurovascular coupling. Physiol Rev. 2006;86(3):1009–31.
101. Kettenmann H, Verkhratsky A. Neuroglia–living nerve glue. Fortschr Neurol Psychiatr. 2011;79(10):588–97.
102. Lynch MA. The multifaceted profile of activated microglia. Mol Neurobiol. 2009;40(2): 139–56.
103. Gomes C, Ferreira R, George J, Sanches R, Rodrigues DI, Goncalves N, et al. Activation of microglial cells triggers a release of brain-derived neurotrophic factor (BDNF) inducing their proliferation in an adenosine A2A receptor-dependent manner: A2A receptor blockade prevents BDNF release and proliferation of microglia. J Neuroinflammation. 2013;10:16.
104. Parkhurst CN, Yang G, Ninan I, Savas JN, Yates JR 3rd, Lafaille JJ, et al. Microglia promote learning-dependent synapse formation through brain-derived neurotrophic factor. Cell. 2013;155(7):1596–609.
105. Pascual O, Ben Achour S, Rostaing P, Triller A, Bessis A. Microglia activation triggers astrocyte-mediated modulation of excitatory neurotransmission. Proc Natl Acad Sci U S A. 2012;109(4):197–205.
106. Rebola N, Simoes AP, Canas PM, Tome AR, Andrade GM, Barry CE, et al. Adenosine A2A receptors control neuroinflammation and consequent hippocampal neuronal dysfunction. J Neurochem. 2011;117(1):100–11.
107. Schafer DP, Lehrman EK, Stevens B. The "quad-partite" synapse: microglia-synapse interactions in the developing and mature CNS. Glia. 2013;61(1):24–36.
108. Scianni M, Antonilli L, Chece G, Cristalli G, Di Castro MA, Limatola C, et al. Fractalkine (CX3CL1) enhances hippocampal N-methyl-D-aspartate receptor (NMDAR) function via D-serine and adenosine receptor type A2 (A2AR) activity. J Neuroinflammation. 2013;10:108.
109. Lingamaneni R, Birch ML, Hemmings HC. Widespread inhibition of sodium channel-dependent glutamate release from isolated nerve terminals by isoflurane and propofol. Anesthesiology. 2001;95(6):1460–6.
110. Parpura V, Heneka MT, Montana V, Oliet SH, Schousboe A, Haydon PG, et al. Glial cells in (patho)physiology. J Neurochem. 2012;121(1):4–27.
111. Lauriat TL, McInnes LA. EAAT2 regulation and splicing: relevance to psychiatric and neurological disorders. Mol Psychiatry. 2007;12(12):1065–78.
112. Jia YF, Choi Y, Ayers-Ringler JR, Biernacka JM, Geske JR, Lindberg DR, et al. Differential SLC1A2 promoter methylation in bipolar disorder with or without addiction. Front Cell Neurosci. 2017;11:217.
113. Poletti S, Locatelli C, Radaelli D, Lorenzi C, Smeraldi E, Colombo C, et al. Effect of early stress on hippocampal gray matter is influenced by a functional polymorphism in EAAT2 in bipolar disorder. Prog Neuro-psychopharmacol Biol Psychiatry. 2014;51:146–52.
114. Castillo M, Kwock L, Courvoisie H, Hooper SR. Proton MR spectroscopy in children with bipolar affective disorder: preliminary observations. AJNR Am J Neuroradiol. 2000;21(5): 832–8.
115. Cecil KM, DelBello MP, Morey R, Strakowski SM. Frontal lobe differences in bipolar disorder as determined by proton MR spectroscopy. Bipolar Disord. 2002;4(6):357–65.
116. Dager SR, Friedman SD, Parow A, Demopulos C, Stoll AL, Lyoo IK, et al. Brain metabolic alterations in medication-free patients with bipolar disorder. Arch Gen Psychiatry. 2004;61(5): 450–8.

117. Kubo H, Nakataki M, Sumitani S, Iga JI, Numata S, Kameoka N, et al. 1H-magnetic resonance spectroscopy study of glutamate-related abnormality in bipolar disorder. J Affect Disord. 2017;208:139–44.
118. Ongur D, Jensen JE, Prescot AP, Stork C, Lundy M, Cohen BM, et al. Abnormal glutamatergic neurotransmission and neuronal-glial interactions in acute mania. Biol Psychiatry. 2008;64(8):718–26.
119. Eastwood SL, Harrison PJ. Decreased mRNA expression of netrin-G1 and netrin-G2 in the temporal lobe in schizophrenia and bipolar disorder. Neuropsychopharmacology. 2008;33(4): 933–45.
120. Eastwood SL, Harrison PJ. Markers of glutamate synaptic transmission and plasticity are increased in the anterior cingulate cortex in bipolar disorder. Biol Psychiatry. 2010;67(11): 1010–6.
121. Uezato A, Meador-Woodruff JH, McCullumsmith RE. Vesicular glutamate transporter mRNA expression in the medial temporal lobe in major depressive disorder, bipolar disorder, and schizophrenia. Bipolar Disord. 2009;11(7):711–25.
122. Fatemi SH, Folsom TD, Thuras PD. Deficits in GABA(B) receptor system in schizophrenia and mood disorders: a postmortem study. Schizophr Res. 2011;128(1-3):37–43.
123. Scarr E, Gray L, Keriakous D, Robinson PJ, Dean B. Increased levels of SNAP-25 and synaptophysin in the dorsolateral prefrontal cortex in bipolar I disorder. Bipolar Disord. 2006;8(2):133–43.
124. Kim HJ, Thayer SA. Lithium increases synapse formation between hippocampal neurons by depleting phosphoinositides. Mol Pharmacol. 2009;75(5):1021–30.
125. Khayachi A, Ase A, Liao C, Kamesh A, Kuhlmann N, Schorova L, et al. Chronic lithium treatment alters the excitatory/inhibitory balance of synaptic networks and reduces mGluR5-PKC signalling in mouse cortical neurons. J Psychiatry Neurosci. 2021;46(3):E402–E14.
126. Zhang Y, Wu S, Xie L, Yu S, Zhang L, Liu C, et al. Ketamine within clinically effective range inhibits glutamate transmission from astrocytes to neurons and disrupts synchronization of astrocytic SICs. Front Cell Neurosci. 2019;13:240.
127. Lazarevic V, Yang Y, Flais I, Svenningsson P. Ketamine decreases neuronally released glutamate via retrograde stimulation of presynaptic adenosine A1 receptors. Mol Psychiatry. 2021;26(12):7425–35.
128. Muller HK, Wegener G, Liebenberg N, Zarate CA Jr, Popoli M, Elfving B. Ketamine regulates the presynaptic release machinery in the hippocampus. J Psychiatr Res. 2013;47(7):892–9.
129. Benes FM, Vincent SL, Todtenkopf M. The density of pyramidal and nonpyramidal neurons in anterior cingulate cortex of schizophrenic and bipolar subjects. Biol Psychiatry. 2001;50(6): 395–406.
130. Brauch RA, Adnan El-Masri M, Parker JC Jr, El-Mallakh RS. Glial cell number and neuron/glial cell ratios in postmortem brains of bipolar individuals. J Affect Disord. 2006;91(1): 87–90.
131. Chana G, Landau S, Beasley C, Everall IP, Cotter D. Two-dimensional assessment of cytoarchitecture in the anterior cingulate cortex in major depressive disorder, bipolar disorder, and schizophrenia: evidence for decreased neuronal somal size and increased neuronal density. Biol Psychiatry. 2003;53(12):1086–98.
132. Rajkowska G, Halaris A, Selemon LD. Reductions in neuronal and glial density characterize the dorsolateral prefrontal cortex in bipolar disorder. Biol Psychiatry. 2001;49(9):741–52.
133. Sakai T, Oshima A, Nozaki Y, Ida I, Haga C, Akiyama H, et al. Changes in density of calcium-binding-protein-immunoreactive GABAergic neurons in prefrontal cortex in schizophrenia and bipolar disorder. Neuropathology. 2008;28(2):143–50.
134. Fatemi SH, Earle JA, McMenomy T. Reduction in Reelin immunoreactivity in hippocampus of subjects with schizophrenia, bipolar disorder and major depression. Mol Psychiatry. 2000;5 (6):654–63, 571.

135. Guidotti A, Auta J, Davis JM, Di-Giorgi-Gerevini V, Dwivedi Y, Grayson DR, et al. Decrease in reelin and glutamic acid decarboxylase67 (GAD67) expression in schizophrenia and bipolar disorder: a postmortem brain study. Arch Gen Psychiatry. 2000;57(11):1061–9.
136. Tobe BTD, Crain AM, Winquist AM, Calabrese B, Makihara H, Zhao WN, et al. Probing the lithium-response pathway in hiPSCs implicates the phosphoregulatory set-point for a cytoskeletal modulator in bipolar pathogenesis. Proc Natl Acad Sci U S A. 2017;114(22):E4462–E71.
137. Pantazopoulos H, Wiseman JT, Markota M, Ehrenfeld L, Berretta S. Decreased numbers of somatostatin-expressing neurons in the amygdala of subjects with bipolar disorder or schizophrenia: relationship to circadian rhythms. Biol Psychiatry. 2017;81(6):536–47.
138. Steullet P, Cabungcal JH, Bukhari SA, Ardelt MI, Pantazopoulos H, Hamati F, et al. The thalamic reticular nucleus in schizophrenia and bipolar disorder: role of parvalbumin-expressing neuron networks and oxidative stress. Mol Psychiatry. 2018;23(10):2057–65.
139. Wang AY, Lohmann KM, Yang CK, Zimmerman EI, Pantazopoulos H, Herring N, et al. Bipolar disorder type 1 and schizophrenia are accompanied by decreased density of parvalbumin- and somatostatin-positive interneurons in the parahippocampal region. Acta Neuropathol. 2011;122(5):615–26.
140. Ishii T, Ishikawa M, Fujimori K, Maeda T, Kushima I, Arioka Y, et al. In vitro modeling of the bipolar disorder and schizophrenia using patient-derived induced pluripotent stem cells with copy number variations of PCDH15 and RELN. eNeuro. 2019;6(5):403.
141. Kim KH, Liu J, Sells Galvin RJ, Dage JL, Egeland JA, Smith RC, et al. Transcriptomic analysis of induced pluripotent stem cells derived from patients with bipolar disorder from an old order amish pedigree. PLoS One. 2015;10(11):e0142693.
142. Mitterauer BJ. Balancing and imbalancing effects of astrocytic receptors in tripartite synapses. Common pathophysiological model of mental disorders and epilepsy. Med Hypotheses. 2015;84(4):315–20.

# Microbiota-Gut-Brain Axis in Major Depression: A New Therapeutic Approach

10

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**Abstract**

Major depression is impacted by the disruption of gut microbiota. Defects in gut microbiota can lead to microbiota-gut-brain axis dysfunction and increased vulnerability to major depression. While traditional chemotherapeutic approaches, such as antidepressant use, produce an overall partial therapeutic effect on depression, the gut microbiome has emerged as an effective target for better therapeutic outcomes. Recent representative studies on the microbiota hypothesis to explore the association between gut pathophysiology and major depression have indicated that restoring gut microbiota and microbiota-gut-brain axis could alleviate depression. We reviewed studies that supported the gut microbiota hypothesis to better understand the pathophysiology of depression; we also explored reports suggesting that gut microbiota restoration is an effective approach for improving depression. These findings indicate that gut microbiota and microbiota-gut-brain axis are appropriate new therapeutic targets for major depression.

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**Keywords**

Major depression · Microbiota · Gut-brain axis · Microbiota-gut-brain axis · Psychobiotics · Probiotics · Fecal microorganism transplantation

## 10.1 Introduction

More than a century ago, Metchnikoff proposed that gut microbiota are fundamental to alleviating mental disorders, including major depression, and supplementation with live microorganisms improves disorders. However, his theory has been largely ignored because of limitations, one of which posits a prevailing biological concept that implicates aberrations in the brain and central nervous system in mental disorders [1–3]. Current therapeutic as well as diagnostic approaches to major depression need to be expanded and consider the complex pathophysiology of the disorder [4–6]. A recent therapeutic approach for major depression includes antidepressant use for chemotherapeutic modification of the contents and density of central neurotransmitters to modulate the activity and function of neural structures, circuitries, and networks in the brain [7, 8]. This central neural system-targeted chemotherapeutic approach, however, has led to high relapse and low remission rates of depression, resulting in a considerable number of treatment-refractory patients worldwide [9–11]. The partial efficacy of brain-targeted therapeutics underscores the urgency to identify novel targets, such as gut microbiota, to treat depression.

Major depression diagnosis and treatment approaches need to be reassessed with consideration of the gut microbiota hypothesis, which carries three components of critical importance [12–14]. The first consideration is the interplay between the gut and brain through bidirectional communication pathways, mainly including neural systems [11, 15–19], metabolic systems [20–22], endocrine system [23–26], and immune system [27–32]. The brain and gut are common neural organs, known as the central and enteric nervous systems, respectively [33, 34]. Crosstalk between the brain and gut, also known as the gut-brain axis, using various peripheral systems, could be a potential biomarker and a novel therapeutic target for major depression. Second, major depression and gut-brain axis disruption are causal factors, and this represents the aberrations in enteric live microorganisms, known as gut microbiota [35–40]. The gut-brain axis is frequently referred to as the microbiota-gut-brain axis, which underscores the potential role of the gut microbiota in regulating emotion and behavior. Third, improving gut microbiota and regulating the microbiota-gut-brain axis may lead to enhanced flexibility and operability in therapeutics for major depression [41–44]. Gut microbiota restoration has emerged as a novel treatment option for depression and includes various methods such as supplementation of live microorganisms, diet regulation, and fecal microorganism transplantation [45]. Taken together, major depression can be viewed as a mental disorder that systemically implicates the gut and brain; gut microbiota may be a plausible biomarker for depression, and gut microbiota restoration can be achieved through

non-chemotherapeutic approaches. The following sections will review the gut microbiota hypothesis in detail, with representative research findings for the aforementioned three premises, to improve understanding of the gut microbiota and microbiota-gut-brain axis as potential treatment targets for major depression.

## 10.2 Major Depression: A Gut-Brain Axis Disorder

The gut-brain axis is a bidirectional pathway that transfers signals between the gut and the brain. It connects the gut and brain through multiple peripheral routes, including the neural, endocrine, metabolic, and immune systems (Fig. 10.1). According to the gut microbiota hypothesis, the gut microbiota regulates the brain through the gut-brain axis, which is also referred to as the microbiota-gut-brain axis, to emphasize the role of the microbiota in regulating the brain and multiple peripheral systems. For example, gut microbiota regulate the growth and functions of the

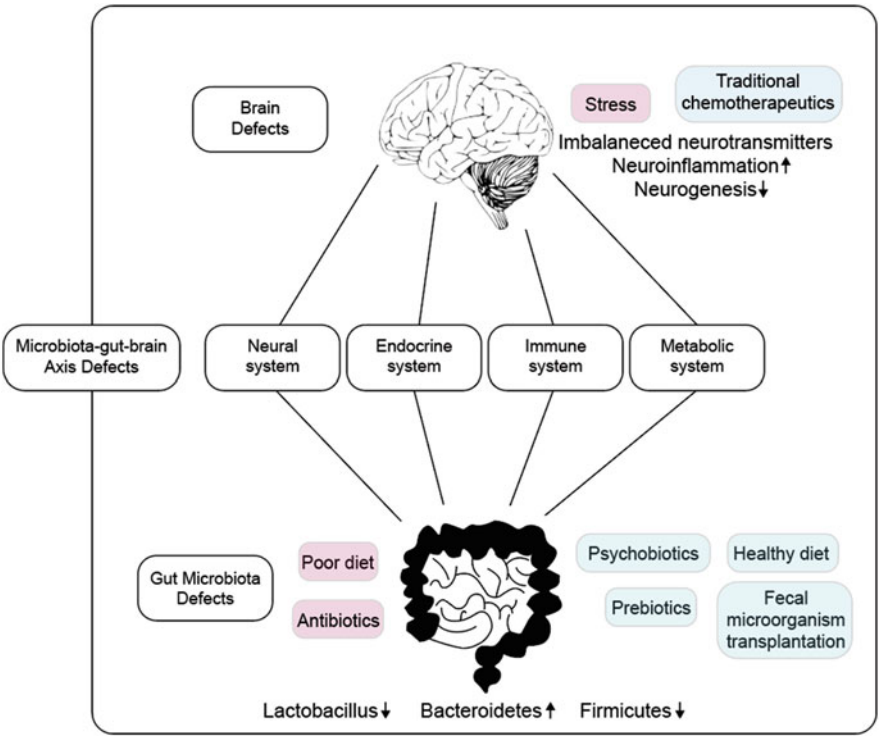


**Fig. 10.1** Microbiota-gut-brain axis defects that contribute to major depression. Microbiota-gut-brain axis defects are major pathophysiology contributors and potential therapeutic targets for major depression. The defects simultaneously impact brain and gut microbiota. The blue boxes indicate therapies that can restore gut microbiota, while the red boxes indicate risk factors for major depression

gut neural systems [35–38], maintain the immune systems [46–48], develop and facilitate maturation of the HPA axis [49–52], and contribute to construction of the blood-brain barrier [53], neurogenesis [54], neuroglia function [55, 56], neurotransmitter synthesis [46–52, 57], myelination [58], and growth of the brain [41, 48, 59, 60]. Hence, gut microbiota are a key component of the gut-brain axis, and restoring the microbiota may ameliorate gut-brain axis dysfunction and aberrations in the neural, endocrine, immune, and metabolic systems.

Major depression is a gut-brain axis disorder with diverse physiological defects that are engraved in the neural, endocrine, immune, and metabolic systems. Physical insults and psychological stress can impair one or more of the multiple peripheral systems that are involved in the gut-brain axis, which may result in gut-brain axis dysfunction and, in turn, major depression [61–63]. The down-top effects (from the gut to the brain) and top-down effects (from the brain to the gut) of the gut-brain axis have been assessed to better understand the extent of the peripheral system involvement [61, 64, 65], both of which emphasize the potential of the gut microbiota in the pathophysiology of major depression. Alterations in the gut microbiota influence emotion and behavior, and changes in the brain also regulate the composition and function of gut microbiota [12–14, 34, 39–41, 66–68]. Some representative studies have suggested mechanisms by which gut microbiota regulate brain function through the gut-brain axis. For the metabolic systems of the gut-brain axis, several studies have indicated that metabolites from carbohydrate and amino acid pathways influence the composition and activity of gut microbiota and even contribute to major depression. Fecal microbiota transplantation from patients with major depression has been reported to induce disturbances in carbohydrate- and amino acid-derived metabolites in mice recipients [21]. The human kynurenine metabolic pathway has been shown to be regulated by *Lactobacillus reuteri* and is also related to major depression [20]. Short-chain fatty acids, such as acetate and butyrate, are gut microbiota-derived mediators in the gut-brain axis [22]. Oral supplementation with short-chain fatty acids ameliorated psychological stress in depressed mouse models [22]. Likewise, for the inflammatory systems of the gut-brain axis, some studies have indicated that the gut microbiota regulate inflammasome-mediated signaling pathways and thus affect emotion and behavior [69]. Caspase-1 is known to cleave pro-interleukin-18 and pro-interleukin-1ß to their mature isoforms in response to stressful stimuli [69]. Wong et al. [69] found that inhibition of caspase-1 increased the abundance of some gut microbiota, such as *Akkermansia* and *Blautia*, and improved stress-associated depressive-like behavior in mice. Oral supplementation with multiple probiotics, including *B. longum* R0175, *L. helveticus* R0052, and *L. plantarum* R1012, affected the composition of pro- and anti-inflammatory cytokines in the hippocampus of depressed mouse models [70]. 5-Hydroxytryptamine is a key metabolite that is involved in major depression and may be regulated by gut microbiota. *Bifidobacterium* regulates the density of 5-hydroxytryptamine and brain-derived neurotrophic factor in the brain [71]. *Clostridium butyricum* increased 5-hydroxytryptamine and glucagon-like peptide-1 concentrations and brain-derived neurotrophic factor expression and reduced depressive-like behavior in mice [72]. Taken together, these findings support the

existence of the gut microbiota-inflammasome-brain axis and the aforementioned gut microbiota-metabolites-brain axis, suggesting the potential role of gut microbiota in regulating the gut-brain axis and associated peripheral systems. Correspondingly, major depression reflects disruption in the microbiota that are crucial for the gut-brain axis and also in the associated peripheral systems.

## 10.3 Gut Microbiota Disruption Is a Stable Hallmark of Major Depression

Major depression can be analyzed from the perspective of gut microbiota disruption. Recent increasing evidence emphasizes the association between disrupted microbiota and depressive phenotypes in both humans and animals. Studies that focused on human subjects have shown that the physiology of gut microbiota significantly differs between patients with major depression and healthy controls. Animal studies have provided evidence for the vital role of gut microbiota in major depression. Corroborating findings from human and animal studies support the gut microbiota hypothesis in major depression, thereby suggesting that gut microbiota disruption is a hallmark of major depression.

### 10.3.1 Associations Between Gut Microbiota Disruption and Major Depression in Human Studies

The gut microbiota hypothesis posits that gut microbiota are involved in the underlying pathophysiology of major depression. Gut microbiota disruption is directly associated with environmental and genetic risks of major depression [39, 40, 67, 68, 73–78]. Clinical trials that focused on major depression have supported a correlation between gut microbiota composition and depressive phenotypes. Gut microbiota composition notably changed in patients with depression [73, 79]. With respect to phyla, the abundance of *Firmicutes* decreased, while that of Bacteroidetes and *Proteobacteria* increased. With respect to family, the abundance of *Prevotellaceae* increased. At the genus level, the abundance of *Faecalibacterium* and *Ruminococcus* decreased, while that of *Prevotella* increased [79, 80]. Furthermore, antibiotic damage may interfere with gut microbiota composition and thus affect an individual's susceptibility to various diseases, including major depression [81–84]. Antibiotics kill both pathogens and beneficial microbiota and lead to gut-brain axis dysfunction. Large-scale human studies have revealed that antibiotic treatment for infectious diseases significantly elevates the risk of psychiatric disorders, including major depression. Psychiatric risk was found to be positively correlated with the time and dose of antibiotic use. The increased risk persisted even 10 years after antibiotic treatment [85, 86]. A study on infants also demonstrated that antibiotic treatment in the first year after birth increases behavioral and psychological problems later in life [87]. In addition to antibiotic use, changes to early microbial exposure, such as during delivery by cesarean section, could hamper the composition of the

microbiota in infants. A cesarean section significantly elevates the alpha diversity of gut microbiota, while reducing the abundance of *Bacteroides*, which regulates intestinal immunity [88]. The elevated alpha diversity of gut microbiota in infants is correlated with a decline in cognitive performance in language and visual reception skills [89]. Assessment of maternal microbiota indicated that cesarean section and breastfeeding were also associated with postpartum depression. Women who underwent cesarean section delivery and discontinued breastfeeding had a higher risk of postpartum depression than controls [90]. A meta-analysis of 532,630 subjects reported an association between cesarean section and postpartum depression with a pooled odds ratio of 1.26 (95% confidence interval, 1.16–1.36) [91]. This association may be due to gastrointestinal dysfunction or infection [91]. However, no human studies have directly examined the causal relationship between gut microbiota and major depression.

### 10.3.2 Associations Between Gut Microbiota Disruption and Depressive-Like Behavior in Animal Studies

Animal studies provide strong evidence for a causal relationship between gut microbiota disruption and depressive-like behavior. The causal relationship has been explored by comparing the gut microbiota of animals with depressive-like behavior and controls, using animal models of depression, such as chronic social defeat stress, maternal separation, and learned helplessness [92, 93]. Recent studies suggest that alterations in gut microbiota are related to changes in depressive-like behavior. To elucidate this relationship, agents that induce microbial alterations have been used, such as prebiotics [94], antibiotics [69], and antidepressant agents [95]. For example, in some chronic stress models, animals were exposed to conditions such as food deprivation, restraint, isolation, and cage tilt for several weeks [71, 96–101]. Animals under chronic stress showed a disrupted gut microbiota, which was associated with chronic stress-induced depressive-like behavior and altered neurotransmitter concentrations [101]. Oral administration of albiflorin [101] and *Lactobacillus* [99] reversed depressive-like behavior of animals. Of note, administration of *L. helveticus* NS8 ameliorated depressive-like behavior and improved cognitive function, and the effects were superior, compared to those with antidepressants such as citalopram [102].

In chronic social defeat stress models, animals are exposed to psychological stress in which members of the same species conflict and attack each other [103]. Mice under chronic social defeat stress exhibited depressive-like behavior such as anhedonia and social avoidance, along with alterations in the microbiota, including a decreased abundance of *Firmicutes* and a decreased ratio of *Firmicutes*/*Bacteroidetes* [104]. Resilience to chronic social defeat stress, on the other hand, was achieved through administration of *Bifidobacterium* [105], indicating that *Bifidobacterium* reduces depressive symptoms and improves stress resilience. With respect to maternal separation models, animals are exposed to early life stress, which is thought to provoke long-lasting psychological vulnerability that leads to the risk of

mental disorders in adulthood [106]. After oral administration of *Bifidobacterium infantis* 35,624, rats exposed to maternal separation exhibited biological profile restoration, such as noradrenaline concentrations in the brain and immune response, as well as improvements in behavior, such as immobility time in the forced swim test [107, 108]. Finally, for the learned helplessness models, animals were exposed to unavoidable shocks and examined for subsequent behavioral tests. The learned helplessness-induced depression model decreased the abundance of gut microbiota, including *Clostridiales incertae sedis* [109] and *Lactobacillaceae* [110]. Of note, consumption of probiotics blended with galactooligosaccharides and polydextrose early in life increased the *Lactobacillus* population and reduced the learned helplessness-induced depressive-like behaviors [111]. Taken together, these studies clearly supported the induced disruption of gut microbiota leading to defective neural function, hampered social behavior, and elevated susceptibility to depression, indicating that gut microbiota disruption is a stable hallmark of major depression.

## 10.4 Gut Microbiota Restoration Alleviates Major Depression

New therapeutics to restore gut microbiota have promising antidepressive effects [40–44]. Several methods to restore gut microbiota have been introduced, including the use of probiotics and fecal microorganism transplantation [112–114]. In this regard, healthy diets and prebiotics have emerged as an alternative therapeutic approach for depression. Importantly, an integrative approach of antidepressant use and probiotic supplementation as an adjuvant helps further advance therapeutics for major depression.

Probiotics are live microorganisms that confer a health benefit to the host when administered in adequate amounts [115]. The beneficial effects are not isolated to the gut, but also reach the gut-brain axis. These probiotics could be conceptualized as psychobiotics to underscore their abilities to improve social behavior and emotion [116]. The psychobiotics that have been studied most frequently belong to lactic acid bacteria, including *Bifidobacterium bifidum* [117, 118], *Lactobacillus casei* [117, 118], and *Lactobacillus helveticus* [102]. Supplementation with psychobiotics improved the gut-brain axis and ameliorated emotional stress in both volunteers and patients with major depression [119]. Daily oral supplementation with probiotic strains alleviated the vulnerability of subjects to mental disorders. A randomized controlled study tested the antidepressant effects of multispecies psychobiotics, including *B. bifidum* W23, *B. lactis* W52, *L. brevis* W63, *L. casei* W56, *L. lactis*, and *L. salivarius* W24, and found that psychobiotic supplementation for 4 weeks significantly improved cognitive reactivity to negative emotions, such as sadness, in patients with major depression, compared with controls that received placebo [120]. Psychobiotic supplementation effectively reduced depression scale scores for both patients with major depression and healthy volunteers. In particular, the psychobiotic effects were more prominent for individuals under 60 years of age than older individuals [121]. However, a more recent study that involved 1349 subjects reported different results. In a study that used psychobiotics, the authors found that

there was an insignificant effect on emotion in healthy subjects but a significant effect in patients with mild-to-moderate depression [122]. In comparison, another study found that there was no significant difference between the psychobiotic (*Bifidobacterium* and *L. helveticus*)-supplemented and placebo-treated groups [123]. These contradictory findings indicate that the psychobiotic effect on major depression might depend on the population and bacterial strains used.

Probiotics, also called psychobiotics, function as adjuvants for antidepressant agents in treating major depression [124]. A clinical trial adopted a combination of *C. butyricum* MIYAIRI 588 and selective serotonin reuptake inhibitors, including duloxetine, escitalopram, fluvoxamine, and sertraline, and found a >50% reduction and a 35% remission rate in 17-item Hamilton depression rating scale scores [125]. Another study found that the combined administration of selective serotonin reuptake inhibitors and probiotics/magnesium orotate formulations, including *L. acidophilus*, *B. bifidum*, and *Streptococcus thermophiles*, significantly improved depression severity scores and quality of life in patients with treatment-refractory major depression [126]. Furthermore, cessation of the probiotic adjuvants led to depression relapse [126]. Consequently, research indicates that probiotics can enhance the therapeutic effect of chemotherapeutic agents on major depression, even in treatment-refractory depression.

Recent studies that evaluated transplanted fecal microorganisms from patients with major depression to stress-naïve animals support the role of gut microbiota in the manifestation of depressive-like behavior [21, 73, 102, 127–130]. After fecal microorganism transplantation, rats exhibited depressive-like behavior, such as anhedonia, in the sucrose consumption test [73]. Fecal microorganism transplantation from patients with major depression led to an increased abundance of Actinobacteria and decreased mobility time in the forced swim and tail suspension tests [21]. Similarly, stress-naïve rats that received microbiota from rats with depressive-like phenotypes demonstrated depressive-like behavior [131]. Rodents that received fecal microorganism transplantation from healthy hosts showed improvements in depressive-like behavior, whereas transplantation from patients with major depression led to depressive phenotypes [132, 133]. However, other studies indicated that probiotic transplantation might not be correlated with a reduction in the incidence of depression [40]. The debate on the efficacy of microorganism transplantation is currently ongoing, and there are limitations to comparing the findings due to discrepancies in bacterial strains, dose, and time of probiotic use.

Contrary to the psychobiotics and fecal microorganism transplantation approaches, both prebiotics and healthy diets have been more recently highlighted for their alternative therapeutic potential for major depression. Prebiotics are defined as substrates that confer a health benefit to the host when selectively utilized by the host microorganisms [134]. Prebiotics not only regulate gut microbiota but also improve social behavior and emotion, acting in a similar manner to psychobiotics. The prebiotic effects may also be achieved through improvements in the gut-brain axis; however, currently there is debate on the efficacy of prebiotics in treating major depression [111, 112, 135, 136]. The most frequently studied types of prebiotics are omega-3 fatty acids, fructose-oligosaccharides, and galactooligosaccharides

[134]. In addition, healthy diets are a hot research topic especially in regard to gut microbiota. Healthy diets improve gut microbiota stability and diversity, thereby leading to psychological and physical well-being [112, 137–139]. Healthy diets increase the abundance of beneficial microbiota and improve cognition and behavior, presumably through the gut-brain axis [114, 137, 139–141]. Healthy diets, such as the Mediterranean diet, are enriched in fermented foods, dietary fiber, and unsaturated fatty acids and contain less food additives, sugar, saturated fatty acids, and refined carbohydrates. Recent studies support dietotherapy for major depression [139]. Furthermore, a recent study combined psychotherapy and dietotherapy to treat patients with panic attacks. In this study, the authors removed sugar-rich foods and increased foods rich in probiotics. This integrative approach ameliorated the anxiety symptoms of the patients and increased the fecal abundance of beneficial microbiota such as *Lactobacillus* [142]. There are various effective methods for restoring gut microbiota and alleviating depression phenotypes, and these can be considered as alternatives or adjuvants to depression therapeutics.

## 10.5 Conclusions

Major depression is a heterogeneous disorder associated with the physiology of the brain, gut, and other peripheral systems. The focus of research on major depression has transitioned from the mind to the brain, to other peripheral systems, to the gut-brain axis, and finally to the microbiota-gut-brain axis. According to the gut microbiota hypothesis, gut microbiota disruption directly induces depressive phenotypes, gut microbiota affects emotion and behavior through the microbiota-gut-brain axis, and microbiota-gut-brain axis disruption is an essential pathophysiology of major depression [12–14, 143–145]. Based on this hypothesis, restoring the gut microbiota and regulating the microbiota-gut-brain axis may be an effective therapeutic approach for major depression. Further, major microbiota restoration methods have been established, including psychobiotics and fecal microorganism transplantation. In particular, the combined use of chemotherapeutic agents and psychobiotic adjuvants broadens the therapeutic boundary for major depression and even improves efficacy in patients with treatment-refractory depression. Therapies that target the gut microbiota and the microbiota-gut-brain axis are expected to progress and become an invaluable approach for treating major depression.

## References

1. Bested AC, Logan AC, Selhub EM. Intestinal microbiota, probiotics and mental health: from Metchnikoff to modern advances: part II–contemporary contextual research. Gut Pathog. 2013;5(1):1–14.
2. Bested AC, Logan AC, Selhub EM. Intestinal microbiota, probiotics and mental health: from Metchnikoff to modern advances: part I–autointoxication revisited. Gut Pathog. 2013;5(1):1–16.

3. Bested AC, Logan AC, Selhub EM. Intestinal microbiota, probiotics and mental health: from Metchnikoff to modern advances: part III–convergence toward clinical trials. Gut Pathog. 2013;5(1):1–13.
4. Kim Y-K, Park S-C. An alternative approach to future diagnostic standards for major depressive disorder. Prog Neuro-Psychopharmacol Biol Psychiatry. 2020;105:110133.
5. Park S-C, Kim Y-K. Challenges and strategies for current classifications of depressive disorders: proposal for future diagnostic standards. In: Major depressive disorder: rethinking and understanding recent discoveries; 2021. p. 103.
6. Kim IB, Park S-C. Machine learning-based definition of symptom clusters and selection of antidepressants for depressive syndrome. Diagnostics. 2021;11(9):1631.
7. Park S-C. Neurogenesis and antidepressant action. Cell Tissue Res. 2019;377(1):95–106.
8. Kim IB, Park S-C. Neural circuitry–neurogenesis coupling model of depression. Int J Mol Sci. 2021;22(5):2468.
9. Garay RP, Zarate CA Jr, Charpeaud T, Citrome L, Correll CU, Hameg A, et al. Investigational drugs in recent clinical trials for treatment-resistant depression. Expert Rev Neurother. 2017;17(6):593–609.
10. Ledford H. Medical research: if depression were cancer. Nature News. 2014;515(7526):182.
11. Chaudhury D, Liu H, Han M-H. Neuronal correlates of depression. Cell Mol Life Sci. 2015;72 (24):4825–48.
12. Smith PA. Brain, meet gut. Nature. 2015;526(7573):312.
13. Mayer EA, Knight R, Mazmanian SK, Cryan JF, Tillisch K. Gut microbes and the brain: paradigm shift in neuroscience. J Neurosci. 2014;34(46):15490–6.
14. Foster JA, Lyte M, Meyer E, Cryan JF. Gut microbiota and brain function: an evolving field in neuroscience. Int J Neuropsychopharmacol. 2016;19(5):pyv114.
15. Liu B, Liu J, Wang M, Zhang Y, Li L. From serotonin to neuroplasticity: evolvement of theories for major depressive disorder. Front Cell Neurosci. 2017;11:305.
16. Serafini G. Neuroplasticity and major depression, the role of modern antidepressant drugs. World J Psychiatry. 2012;2(3):49.
17. Lener MS, Niciu MJ, Ballard ED, Park M, Park LT, Nugent AC, et al. Glutamate and gamma-aminobutyric acid systems in the pathophysiology of major depression and antidepressant response to ketamine. Biol Psychiatry. 2017;81(10):886–97.
18. Pytka K, Dziubina A, Młyniec K, Dziedziczak A, Żmudzka E, Furgała A, et al. The role of glutamatergic, GABA-ergic, and cholinergic receptors in depression and antidepressant-like effect. Pharmacol Rep. 2016;68(2):443–50.
19. Murrough JW, Abdallah CG, Mathew SJ. Targeting glutamate signalling in depression: progress and prospects. Nat Rev Drug Discov. 2017;16(7):472–86.
20. Xie R, Jiang P, Lin L, Jiang J, Yu B, Rao J, et al. Oral treatment with Lactobacillus reuteri attenuates depressive-like behaviors and serotonin metabolism alterations induced by chronic social defeat stress. J Psychiatr Res. 2020;122:70–8.
21. Zheng P, Zeng B, Zhou C, Liu M, Fang Z, Xu X, et al. Gut microbiome remodeling induces depressive-like behaviors through a pathway mediated by the host's metabolism. Mol Psychiatry. 2016;21(6):786–96.
22. van de Wouw M, Boehme M, Lyte JM, Wiley N, Strain C, O'Sullivan O, et al. Short-chain fatty acids: microbial metabolites that alleviate stress-induced brain–gut axis alterations. J Physiol. 2018;596(20):4923–44.
23. Belmaker R, Agam G. Major depressive disorder. N Engl J Med. 2008;358(1):55–68.
24. Lima-Ojeda JM, Rupprecht R, Baghai TC. "I am I and my bacterial circumstances": linking gut microbiome, neurodevelopment, and depression. Front Psych. 2017;8:153.
25. Barden N. Implication of the hypothalamic–pituitary–adrenal axis in the physiopathology of depression. J Psychiatry Neurosci. 2004;29(3):185.
26. Juruena MF, Cleare AJ, Pariante CM. The hypothalamic pituitary adrenal axis, glucocorticoid receptor function and relevance to depression. Braz J Psychiatry. 2004;26:189–201.

27. Maes M, Leonard B, Myint A, Kubera M, Verkerk R. The new '5-HT'hypothesis of depression: cell-mediated immune activation induces indoleamine 2, 3-dioxygenase, which leads to lower plasma tryptophan and an increased synthesis of detrimental tryptophan catabolites (TRYCATs), both of which contribute to the onset of depression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2011;35(3):702–21.
28. Singhal G, Baune BT. Microglia: an interface between the loss of neuroplasticity and depression. Front Cell Neurosci. 2017;11:270.
29. O'Brien SM, Scott LV, Dinan TG. Cytokines: abnormalities in major depression and implications for pharmacological treatment. Hum Psychopharmacol Clin Exp. 2004;19(6): 397–403.
30. Wichers M, Maes M. The psychoneuroimmuno-pathophysiology of cytokine-induced depression in humans. Int J Neuropsychopharmacol. 2002;5(4):375–88.
31. Leonard BE. Inflammation and depression: a causal or coincidental link to the pathophysiology? Acta Neuropsychiatr. 2018;30(1):1–16.
32. Haroon E, Miller AH. Inflammation effects on brain glutamate in depression: mechanistic considerations and treatment implications. Curr Top Behav Neurosci. 2017;31:173–98.
33. Rao M, Gershon MD. The bowel and beyond: the enteric nervous system in neurological disorders. Nat Rev Gastroenterol Hepatol. 2016;13(9):517–28.
34. Liang S, Wang T, Hu X, Li W, Jin F, Wang L. Microorganism and behavior and psychiatric disorders. Adv Psychol Sci. 2012;20:75–97.
35. O'Hara AM, Shanahan F. The gut flora as a forgotten organ. EMBO Rep. 2006;7(7):688–93.
36. Lyte M. The microbial organ in the gut as a driver of homeostasis and disease. Med Hypotheses. 2010;74(4):634–8.
37. Avetisyan M, Schill EM, Heuckeroth RO. Building a second brain in the bowel. J Clin Invest. 2015;125(3):899–907.
38. Knight R, Callewaert C, Marotz C, Hyde ER, Debelius JW, McDonald D, et al. The microbiome and human biology. Annu Rev Genomics Hum Genet. 2017;18:65–86.
39. Kelly JR, Clarke G, Cryan JF, Dinan TG. Brain-gut-microbiota axis: challenges for translation in psychiatry. Ann Epidemiol. 2016;26(5):366–72.
40. Rieder R, Wisniewski PJ, Alderman BL, Campbell SC. Microbes and mental health: a review. Brain Behav Immun. 2017;66:9–17.
41. Kennedy PJ, Murphy AB, Cryan JF, Ross PR, Dinan TG, Stanton C. Microbiome in brain function and mental health. Trends Food Sci Technol. 2016;57:289–301.
42. Grenham S, Clarke G, Cryan JF, Dinan TG. Brain–gut–microbe communication in health and disease. Front Physiol. 2011;2:94.
43. Clemente JC, Ursell LK, Parfrey LW, Knight R. The impact of the gut microbiota on human health: an integrative view. Cell. 2012;148(6):1258–70.
44. Shanahan F. The hygiene hypothesis and darwinian medicine. Basel: Birkhäuser; 2009.
45. Fond GB, Lagier J-C, Honore S, Lancon C, Korchia T, Verville P-LSD, et al. Microbiota-orientated treatments for major depression and schizophrenia. Nutrients. 2020;12(4):1024.
46. Honda K, Littman DR. The microbiota in adaptive immune homeostasis and disease. Nature. 2016;535(7610):75–84.
47. Thaiss CA, Levy M, Suez J, Elinav E. The interplay between the innate immune system and the microbiota. Curr Opin Immunol. 2014;26:41–8.
48. Kim S, Kim H, Yim YS, Ha S, Atarashi K, Tan TG, et al. Maternal gut bacteria promote neurodevelopmental abnormalities in mouse offspring. Nature. 2017;549(7673):528–32.
49. Microbiome SN. HPA axis and production of endocrine hormones in the gut. Microbial endocrinology: the microbiota-gut-brain axis in health and disease. Springer; 2014. p. 177–94.
50. Gareau MG, Jury J, MacQueen G, Sherman PM, Perdue MH. Probiotic treatment of rat pups normalises corticosterone release and ameliorates colonic dysfunction induced by maternal separation. Gut. 2007;56(11):1522–8.
51. Eutamene H, Bueno L. Role of probiotics in correcting abnormalities of colonic flora induced by stress. Gut. 2007;56(11):1495–7.

52. Sudo N, Chida Y, Aiba Y, Sonoda J, Oyama N, Yu XN, et al. Postnatal microbial colonization programs the hypothalamic–pituitary–adrenal system for stress response in mice. J Physiol. 2004;558(1):263–75.
53. Bien-Ly N, Watts RJ. The blood-brain barrier's gut check. Sci Transl Med. 2014;6(263): 263fs46-fs46.
54. Ogbonnaya ES, Clarke G, Shanahan F, Dinan TG, Cryan JF, O'Leary OF. Adult hippocampal neurogenesis is regulated by the microbiome. Biol Psychiatry. 2015;78(4):e7–9.
55. Castillo-Ruiz A, Mosley M, George AJ, Mussaji LF, Fullerton EF, Ruszkowski EM, et al. The microbiota influences cell death and microglial colonization in the perinatal mouse brain. Brain Behav Immun. 2018;67:218–29.
56. Erny D, de Angelis ALH, Jaitin D, Wieghofer P, Staszewski O, David E, et al. Host microbiota constantly control maturation and function of microglia in the CNS. Nat Neurosci. 2015;18(7): 965–77.
57. O'Mahony SM, Clarke G, Borre Y, Dinan T, Cryan J. Serotonin, tryptophan metabolism and the brain-gut-microbiome axis. Behav Brain Res. 2015;277:32–48.
58. Hoban AE, Stilling RM, Ryan FJ, Shanahan F, Dinan TG, Claesson MJ, et al. Regulation of prefrontal cortex myelination by the microbiota. Transl Psychiatry. 2016;6(4):e774-e.
59. Borre YE, O'Keeffe GW, Clarke G, Stanton C, Dinan TG, Cryan JF. Microbiota and neurodevelopmental windows: implications for brain disorders. Trends Mol Med. 2014;20 (9):509–18.
60. Heijtz RD, Wang S, Anuar F, Qian Y, Björkholm B, Samuelsson A, et al. Normal gut microbiota modulates brain development and behavior. Proc Natl Acad Sci. 2011;108(7): 3047–52.
61. Wilhelmsen I. Brain-gut axis as an example of the bio-psycho-social model. Gut. 2000;47 (Suppl 4):iv5–7.
62. O'Mahony SM, Hyland NP, Dinan TG, Cryan JF. Maternal separation as a model of brain–gut axis dysfunction. Psychopharmacology. 2011;214(1):71–88.
63. Scott LV, Clarke G, Dinan TG. The brain-gut axis: a target for treating stress-related disorders. Mod Trends Pharmacopsychiatry. 2013;28:90–9.
64. Neufeld K-A, Foster JA. Effects of gut microbiota on the brain: implications for psychiatry. J Psychiatry Neurosci. 2009;34(3):230.
65. Collins SM, Bercik P. The relationship between intestinal microbiota and the central nervous system in normal gastrointestinal function and disease. Gastroenterology. 2009;136(6): 2003–14.
66. Mayer EA, Tillisch K, Gupta A. Gut/brain axis and the microbiota. J Clin Invest. 2015;125(3): 926–38.
67. Yarandi SS, Peterson DA, Treisman GJ, Moran TH, Pasricha PJ. Modulatory effects of gut microbiota on the central nervous system: how gut could play a role in neuropsychiatric health and diseases. J Neurogastroenterol Motilit. 2016;22(2):201.
68. Fond G, Boukouaci W, Chevalier G, Regnault A, Eberl G, Hamdani N, et al. The "psychomicrobiotic": targeting microbiota in major psychiatric disorders: a systematic review. Pathol Biol. 2015;63(1):35–42.
69. Wong M-L, Inserra A, Lewis M, Mastronardi CA, Leong L, Choo J, et al. Inflammasome signaling affects anxiety-and depressive-like behavior and gut microbiome composition. Mol Psychiatry. 2016;21(6):797–805.
70. Li B, Guo K, Zeng L, Zeng B, Huo R, Luo Y, et al. Metabolite identification in fecal microbiota transplantation mouse livers and combined proteomics with chronic unpredictive mild stress mouse livers. Transl Psychiatry. 2018;8(1):1–12.
71. Sun L, Zhang H, Cao Y, Wang C, Zhao C, Wang H, et al. Fluoxetine ameliorates dysbiosis in a depression model induced by chronic unpredicted mild stress in mice. Int J Med Sci. 2019;16 (9):1260.

72. Sun J, Wang F, Hu X, Yang C, Xu H, Yao Y, et al. Clostridium butyricum attenuates chronic unpredictable mild stress-induced depressive-like behavior in mice via the gut-brain axis. J Agric Food Chem. 2018;66(31):8415–21.
73. Kelly JR, Borre Y, O'Brien C, Patterson E, El Aidy S, Deane J, et al. Transferring the blues: depression-associated gut microbiota induces neurobehavioural changes in the rat. J Psychiatr Res. 2016;82:109–18.
74. Luna RA, Foster JA. Gut brain axis: diet microbiota interactions and implications for modulation of anxiety and depression. Curr Opin Biotechnol. 2015;32:35–41.
75. Maqsood R, Stone TW. The gut-brain axis, BDNF, NMDA and CNS disorders. Neurochem Res. 2016;41(11):2819–35.
76. Farmer AD, Randall HA, Aziz Q. It's a gut feeling: how the gut microbiota affects the state of mind. J Physiol. 2014;592(14):2981–8.
77. Kennedy PJ, Cryan JF, Dinan TG, Clarke G. Kynurenine pathway metabolism and the microbiota-gut-brain axis. Neuropharmacology. 2017;112:399–412.
78. Cryan JF, Dinan TG. Mind-altering microorganisms: the impact of the gut microbiota on brain and behaviour. Nat Rev Neurosci. 2012;13(10):701–12.
79. Jiang H, Ling Z, Zhang Y, Mao H, Ma Z, Yin Y, et al. Altered fecal microbiota composition in patients with major depressive disorder. Brain Behav Immun. 2015;48:186–94.
80. Liu Y, Zhang L, Wang X, Wang Z, Zhang J, Jiang R, et al. Similar fecal microbiota signatures in patients with diarrhea-predominant irritable bowel syndrome and patients with depression. Clin Gastroenterol Hepatol. 2016;14(11):1602–1611.e5.
81. Hu X, Wang T, Liang S, Li W, Wu X, Jin F. Antibiotic-induced imbalances in gut microbiota aggravates cholesterol accumulation and liver injuries in rats fed a high-cholesterol diet. Appl Microbiol Biotechnol. 2015;99(21):9111–22.
82. Fröhlich EE, Farzi A, Mayerhofer R, Reichmann F, Jačan A, Wagner B, et al. Cognitive impairment by antibiotic-induced gut dysbiosis: analysis of gut microbiota-brain communication. Brain Behav Immun. 2016;56:140–55.
83. Bercik P, Collins SM. The effects of inflammation, infection and antibiotics on the microbiota-gut-brain axis. In: Microbial endocrinology: the microbiota-gut-brain axis in health and disease; 2014. p. 279–89.
84. Wang T, Hu X, Liang S, Li W, Wu X, Wang L, et al. Lactobacillus fermentum NS9 restores the antibiotic induced physiological and psychological abnormalities in rats. Benefic Microbes. 2015;6(5):707–17.
85. Lurie I, Yang Y-X, Haynes K, Mamtani R, Boursi B. Antibiotic exposure and the risk for depression, anxiety, or psychosis: a nested case-control study. J Clin Psychiatry. 2015;76(11):1522–8.
86. Köhler O, Petersen L, Mors O, Mortensen P, Yolken RH, Gasse C, et al. Infections and exposure to anti-infective agents and the risk of severe mental disorders: a nationwide study. Acta Psychiatr Scand. 2017;135(2):97–105.
87. Slykerman RF, Thompson J, Waldie KE, Murphy R, Wall C, Mitchell EA. Antibiotics in the first year of life and subsequent neurocognitive outcomes. Acta Paediatr. 2017;106(1):87–94.
88. Bokulich N, Chung J, Battaglia T, Henderson N, Jay M, Li H, et al. Antibiotics, birth mode, and diet shape microbiome maturation during early life. Sci Transl Med. 2016;8:343ra82.
89. Carlson AL, Xia K, Azcarate-Peril MA, Goldman BD, Ahn M, Styner MA, et al. Infant gut microbiome associated with cognitive development. Biol Psychiatry. 2018;83(2):148–59.
90. Nam JY, Choi Y, Kim J, Cho KH, Park E-C. The synergistic effect of breastfeeding discontinuation and cesarean section delivery on postpartum depression: a nationwide population-based cohort study in Korea. J Affect Disord. 2017;218:53–8.
91. Xu H, Ding Y, Ma Y, Xin X, Zhang D. Cesarean section and risk of postpartum depression: a meta-analysis. J Psychosom Res. 2017;97:118–26.
92. Deisseroth KA, Tye KM, Warden MR. Non-human animal models of depression and methods of use thereof. Google Patents; 2015.

93. Czéh B, Fuchs E, Wiborg O, Simon M. Animal models of major depression and their clinical implications. Prog Neuro-Psychopharmacol Biol Psychiatry. 2016;64:293–310.
94. Maehata H, Kobayashi Y, Mitsuyama E, Kawase T, Kuhara T, Xiao J-Z, et al. Heat-killed lactobacillus helveticus strain MCC1848 confers resilience to anxiety or depression-like symptoms caused by subchronic social defeat stress in mice. Biosci Biotechnol Biochem. 2019;83(7):1239–47.
95. Cheng D, Chang H, Ma S, Guo J, She G, Zhang F, et al. Tiansi liquid modulates gut microbiota composition and tryptophan–kynurenine metabolism in rats with hydrocortisone-induced depression. Molecules. 2018;23(11):2832.
96. Rincel M, Aubert P, Chevalier J, Grohard P-A, Basso L, de Oliveira CM, et al. Multi-hit early life adversity affects gut microbiota, brain and behavior in a sex-dependent manner. Brain Behav Immun. 2019;80:179–92.
97. Zhang Y, Huang R, Cheng M, Wang L, Chao J, Li J, et al. Gut microbiota from NLRP3-deficient mice ameliorates depressive-like behaviors by regulating astrocyte dysfunction via circHIPK2. Microbiome. 2019;7(1):1–16.
98. Tian P, Wang G, Zhao J, Zhang H, Chen W. Bifidobacterium with the role of 5-hydroxytryptophan synthesis regulation alleviates the symptom of depression and related microbiota dysbiosis. J Nutr Biochem. 2019;66:43–51.
99. Marin IA, Goertz JE, Ren T, Rich SS, Onengut-Gumuscu S, Farber E, et al. Microbiota alteration is associated with the development of stress-induced despair behavior. Sci Rep. 2017;7(1):1–10.
100. Li Y, Peng Y, Ma P, Yang H, Xiong H, Wang M, et al. Antidepressant-like effects of Cistanche tubulosa extract on chronic unpredictable stress rats through restoration of gut microbiota homeostasis. Front Pharmacol. 2018;9:967.
101. Jianguo L, Xueyang J, Cui W, Changxin W, Xuemei Q. Altered gut metabolome contributes to depression-like behaviors in rats exposed to chronic unpredictable mild stress. Transl Psychiatry. 2019;9(1):1–14.
102. Liang S, Wang T, Hu X, Luo J, Li W, Wu X, et al. Administration of Lactobacillus helveticus NS8 improves behavioral, cognitive, and biochemical aberrations caused by chronic restraint stress. Neuroscience. 2015;310:561–77.
103. Valleau JC, Sullivan EL. The impact of leptin on perinatal development and psychopathology. J Chem Neuroanat. 2014;61-62:221–32.
104. Zhang J, Yao W, Dong C, Yang C, Ren Q, Ma M, et al. Blockade of interleukin-6 receptor in the periphery promotes rapid and sustained antidepressant actions: a possible role of gut–microbiota–brain axis. Transl Psychiatry. 2017;7(5):e1138-e.
105. Yang C, Fujita Y, Ren Q, Ma M, Dong C, Hashimoto K. Bifidobacterium in the gut microbiota confer resilience to chronic social defeat stress in mice. Sci Rep. 2017;7(1):1–7.
106. Kendler KS, Gardner CO, Prescott CA. Toward a comprehensive developmental model for major depression in women. Am J Psychiatr. 2002;159(7):1133–45.
107. O'Mahony SM, Marchesi JR, Scully P, Codling C, Ceolho A-M, Quigley EM, et al. Early life stress alters behavior, immunity, and microbiota in rats: implications for irritable bowel syndrome and psychiatric illnesses. Biol Psychiatry. 2009;65(3):263–7.
108. Desbonnet L, Garrett L, Clarke G, Kiely B, Cryan JF, Dinan TG. Effects of the probiotic Bifidobacterium infantis in the maternal separation model of depression. Neuroscience. 2010;170(4):1179–88.
109. Takajo T, Tomita K, Tsuchihashi H, Enomoto S, Tanichi M, Toda H, et al. Depression promotes the onset of irritable bowel syndrome through unique dysbiosis in rats. Gut Liver. 2019;13(3):325.
110. Zhang K, Fujita Y, Chang L, Qu Y, Pu Y, Wang S, et al. Abnormal composition of gut microbiota is associated with resilience versus susceptibility to inescapable electric stress. Transl Psychiatry. 2019;9(1):1–9.
111. Mika A, Day HE, Martinez A, Rumian NL, Greenwood BN, Chichlowski M, et al. Early life diets with prebiotics and bioactive milk fractions attenuate the impact of stress on learned

helplessness behaviours and alter gene expression within neural circuits important for stress resistance. Eur J Neurosci. 2017;45(3):342–57.
112. Liu X, Cao S, Zhang X. Modulation of gut microbiota–brain axis by probiotics, prebiotics, and diet. J Agric Food Chem. 2015;63(36):7885–95.
113. Cammarota G, Ianiro G, Bibbo S, Gasbarrini A. Gut microbiota modulation: probiotics, antibiotics or fecal microbiota transplantation? Intern Emerg Med. 2014;9(4):365–73.
114. Marques TM, Cryan JF, Shanahan F, Fitzgerald GF, Ross RP, Dinan TG, et al. Gut microbiota modulation and implications for host health: dietary strategies to influence the gut–brain axis. Innovative Food Sci Emerg Technol. 2014;22:239–47.
115. Hotel ACP, Cordoba A. Health and nutritional properties of probiotics in food including powder milk with live lactic acid bacteria. Prevention. 2001;5(1):1–10.
116. Dinan TG, Stanton C, Cryan JF. Psychobiotics: a novel class of psychotropic. Biol Psychiatry. 2013;74(10):720–6.
117. Akkasheh G, Kashani-Poor Z, Tajabadi-Ebrahimi M, Jafari P, Akbari H, Taghizadeh M, et al. Clinical and metabolic response to probiotic administration in patients with major depressive disorder: a randomized, double-blind, placebo-controlled trial. Nutrition. 2016;32(3):315–20.
118. Abildgaard A, Elfving B, Hokland M, Wegener G, Lund S. Probiotic treatment reduces depressive-like behaviour in rats independently of diet. Psychoneuroendocrinology. 2017;79:40–8.
119. Pirbaglou M, Katz J, de Souza RJ, Stearns JC, Motamed M, Ritvo P. Probiotic supplementation can positively affect anxiety and depressive symptoms: a systematic review of randomized controlled trials. Nutr Res. 2016;36(9):889–98.
120. Steenbergen L, Sellaro R, van Hemert S, Bosch JA, Colzato LS. A randomized controlled trial to test the effect of multispecies probiotics on cognitive reactivity to sad mood. Brain Behav Immun. 2015;48:258–64.
121. Huang R, Wang K, Hu J. Effect of probiotics on depression: a systematic review and meta-analysis of randomized controlled trials. Nutrients. 2016;8(8):483.
122. Ng QX, Peters C, Ho CYX, Lim DY, Yeo W-S. A meta-analysis of the use of probiotics to alleviate depressive symptoms. J Affect Disord. 2018;228:13–9.
123. Romijn AR, Rucklidge JJ, Kuijer RG, Frampton C. A double-blind, randomized, placebo-controlled trial of lactobacillus helveticus and Bifidobacterium longum for the symptoms of depression. Aust N Z J Psychiatry. 2017;51(8):810–21.
124. VlainiH J, Suran J, Letizia Vukorep A. Probiotics as an adjuvant therapy in major depressive disorder. Curr Neuropharmacol. 2016;14(8):952–8.
125. Miyaoka T, Kanayama M, Wake R, Hashioka S, Hayashida M, Nagahama M, et al. Clostridium butyricum MIYAIRI 588 as adjunctive therapy for treatment-resistant major depressive disorder: a prospective open-label trial. Clin Neuropharmacol. 2018;41(5):151–5.
126. Bambling M, Edwards SC, Hall S, Vitetta L. A combination of probiotics and magnesium orotate attenuate depression in a small SSRI resistant cohort: an intestinal anti-inflammatory response is suggested. Inflammopharmacology. 2017;25(2):271–4.
127. Gacias M, Gaspari S, Santos P-MG, Tamburini S, Andrade M, Zhang F, et al. Microbiota-driven transcriptional changes in prefrontal cortex override genetic differences in social behavior. eLife. 2016;5:e13442.
128. Bravo JA, Forsythe P, Chew MV, Escaravage E, Savignac HM, Dinan TG, et al. Ingestion of Lactobacillus strain regulates emotional behavior and central GABA receptor expression in a mouse via the vagus nerve. Proc Natl Acad Sci. 2011;108(38):16050–5.
129. Campos AC, Rocha NP, Nicoli JR, Vieira LQ, Teixeira MM, Teixeira AL. Absence of gut microbiota influences lipopolysaccharide-induced behavioral changes in mice. Behav Brain Res. 2016;312:186–94.
130. Savignac H, Kiely B, Dinan T, Cryan J. Bifidobacteria exert strain-specific effects on stress-related behavior and physiology in BALB/c mice. Neurogastroenterol Motil. 2014;26(11):1615–27.

131. Pearson-Leary J, Zhao C, Bittinger K, Eacret D, Luz S, Vigderman AS, et al. The gut microbiome regulates the increases in depressive-type behaviors and in inflammatory processes in the ventral hippocampus of stress vulnerable rats. Mol Psychiatry. 2020;25(5): 1068–79.
132. Koopman M, El Aidy S. Depressed gut? The microbiota-diet-inflammation trialogue in depression. Curr Opin Psychiatry. 2017;30(5):369–77.
133. Mohajeri MH, La Fata G, Steinert RE, Weber P. Relationship between the gut microbiome and brain function. Nutr Rev. 2018;76(7):481–96.
134. Gibson GR, Hutkins R, Sanders ME, Prescott SL, Reimer RA, Salminen SJ, et al. Expert consensus document: the International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of prebiotics. Nat Rev Gastroenterol Hepatol. 2017;14(8):491–502.
135. Schmidt K, Cowen PJ, Harmer CJ, Tzortzis G, Errington S, Burnet PW. Prebiotic intake reduces the waking cortisol response and alters emotional bias in healthy volunteers. Psychopharmacology. 2015;232(10):1793–801.
136. Robertson RC, Oriach CS, Murphy K, Moloney GM, Cryan JF, Dinan TG, et al. Omega-3 polyunsaturated fatty acids critically regulate behaviour and gut microbiota development in adolescence and adulthood. Brain Behav Immun. 2017;59:21–37.
137. Murphy T, Dias GP, Thuret S. Effects of diet on brain plasticity in animal and human studies: mind the gap. Neural Plast. 2014;2014:563160.
138. Heiman ML, Greenway FL. A healthy gastrointestinal microbiome is dependent on dietary diversity. Mol Metab. 2016;5(5):317–20.
139. Dash S, Clarke G, Berk M, Jacka FN. The gut microbiome and diet in psychiatry: focus on depression. Curr Opin Psychiatry. 2015;28(1):1–6.
140. Sandhu KV, Sherwin E, Schellekens H, Stanton C, Dinan TG, Cryan JF. Feeding the microbiota-gut-brain axis: diet, microbiome, and neuropsychiatry. Transl Res. 2017;179: 223–44.
141. Oriach CS, Robertson RC, Stanton C, Cryan JF, Dinan TG. Food for thought: the role of nutrition in the microbiota-gut–brain axis. Clin Nut Exper. 2016;6:25–38.
142. Schnorr SL, Bachner HA. Focus: microbiome: integrative therapies in anxiety treatment with special emphasis on the gut microbiome. Yale J Biol Med. 2016;89(3):397.
143. Dinan TG, Stilling RM, Stanton C, Cryan JF. Collective unconscious: how gut microbes shape human behavior. J Psychiatr Res. 2015;63:1–9.
144. Parashar A, Udayabanu M. Gut microbiota regulates key modulators of social behavior. Eur Neuropsychopharmacol. 2016;26(1):78–91.
145. Mu C, Yang Y, Zhu W. Gut microbiota: the brain peacekeeper. Front Microbiol. 2016;7:345.

# PTSD, Immune System, and Inflammation

11

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**Abstract**

Posttraumatic stress disorder (PTSD) is a severe trauma and stress-related disorder associated with different somatic comorbidities, especially cardiovascular and metabolic disorders, and with chronic low-grade inflammation. Altered balance of the hypothalamic-pituitary-adrenal (HPA) axis, cytokines and chemokines, C-reactive protein, oxidative stress markers, kynurenine pathways, and gut microbiota might be involved in the alterations of certain brain regions regulating fear conditioning and memory processes, that are all altered in PTSD. In addition to the HPA axis, the gut microbiota maintains the balance and interaction of the

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immune, CNS, and endocrine pathways forming the gut-brain axis. Disbalance in the HPA axis, gut-brain axis, oxidative stress pathways and kynurenine pathways, altered immune signaling and disrupted homeostasis, as well as the association of the PTSD with the inflammation and disrupted cognition support the search for novel strategies for treatment of PTSD. Besides potential anti-inflammatory treatment, dietary interventions or the use of beneficial bacteria, such as probiotics, can potentially improve the composition and the function of the bacterial community in the gut. Therefore, bacterial supplements and controlled dietary changes, with exercise, might have beneficial effects on the psychological and cognitive functions in patients with PTSD. These new treatments should be aimed to attenuate inflammatory processes and consequently to reduce PTSD symptoms but also to improve cognition and reduce cardio-metabolic disorders associated so frequently with PTSD.

**Keywords**

Brain-gut axis · Cardiovascular disease · Chemokines · CRP · Kynurenine pathway · Cytokines · HPA axis · Inflammation · Immune system · Oxidative stress · PTSD

## Abbreviations

| | |
|---|---|
| 3-HAA | 3-Hydroxy-anthranilic acid |
| 3-HK | 3-Hydroxy-kynurenine |
| 4-HNE | 4-Hydroxy-2-nonenal |
| 8-OHdG | 8-Hydroxy-2′-deoxyguanosine |
| ACTH | Adrenocorticotropic hormone |
| AIM2 | Absent in melanoma 2 |
| BBB | Blood-brain barrier |
| BDNF | Brain-derived neurotrophic factor |
| BMI | Body mass index |
| CAPS | Clinician administered PTSD scale |
| CNS | Central nervous system |
| CRH | Corticotrophin-releasing hormone |
| CRP | C-reactive protein |
| CSF | Cerebrospinal fluid |
| CVD | Cardiovascular disease |
| DST | Dexamethasone suppression test |
| ELISA | Enzyme-linked immunosorbent assay |
| FDG | Fluorodeoxyglucose |
| GABA | Gamma-aminobutyric acid |
| GPx | Glutathione peroxidase |
| HPA | Hypothalamic pituitary-adrenal axis |
| IDO | Indoleamine 2,3-dioxygenase |

| | |
|---|---|
| IFN-γ | Interferon gamma |
| IgA | Immunoglobulin A |
| IL | Interleukin |
| KAT | Kynurenine aminotransferase |
| KMO | Kynurenine 3-monooxygenase |
| KYN | Kynurenine |
| KYNA | Kynurenic acid |
| LD | Linkage disequilibrium |
| MDA | Malondialdehyde |
| MDD | Major depressive disorder |
| MetS | Metabolic syndrome |
| NAD+ | Nicotinamide adenine dinucleotide |
| NF-κB | Nuclear factor-κB |
| NMDA | *N*-Methyl-D-aspartate |
| *NR3C1-1F* | Glucocorticoid receptor gene exon 1F |
| PC | Phosphatidylcholine |
| PE | Phosphatidylethanolamine |
| PET | Positron emission tomography |
| PTSD | Posttraumatic stress disorder |
| QUIN | Quinolinic acid |
| RAS | Renin-angiotensin system |
| ROS | Reactive oxidative species |
| SNS | Sympathetic nervous system |
| SOD | Superoxide dismutase |
| TBI | Traumatic brain injury |
| TDO | Tryptophan 2,3-dioxygenase |
| TGF-β | Transforming growth factor beta |
| TNF-α | Tumor necrosis factor alpha |
| Treg | Regulatory T cells |

## 11.1 Introduction

Posttraumatic stress disorder (PTSD) is a severe mental disorder that develops in some, but not all individuals who have witnessed or have been exposed to traumatic and stressful events. It affects mental and physical quality of life of patients and their families and is frequently associated with different somatic comorbidities [1]. Although it is a stress-related disorder that affects primarily the brain and the stress circuits, PTSD is believed to be a systemic illness affecting all organ systems [2, 3]. PTSD and trauma exposure are frequently associated with chronic low-grade inflammation [4–9]. Immunological alterations lead to the long-term health consequences [10], while exposure to trauma results in the stimulation of the hypothalamic pituitary-adrenal (HPA) axis and activation of the immune system, with the subsequent release of the pro-inflammatory cytokines [11]. PTSD is a

highly heterogeneous disorder, and it should be acknowledged that not all studies have reported increased inflammation [12].

## 11.2 The Hypothalamic-Pituitary-Adrenal Axis and Inflammation in PTSD

The HPA axis is dysregulated in PTSD [3, 10, 13–16]. It is a main stress system which responds to different stressors and traumas first by the activation of the HPA axis in the interaction with the sympathetic nervous system (SNS). After an acute stress, corticotrophin-releasing hormone (CRH) is released from the paraventricular nucleus of the hypothalamus into the portal blood that stimulates corticotropic cells in the anterior and intermediate lobes of the pituitary gland to secrete proopiomelanocortin, which releases, among other active peptides, also adrenocorticotropic hormone (ACTH). ACTH is released in the circulation and stimulates the synthesis and the release of glucocorticoid hormones from the adrenal cortex. The balance of the HPA axis, with a decrease of the HPA activation and reduced release of CRH, ACTH, and cortisol, is achieved by the negative feedback of cortisol that self-regulate the secretion of CRH, arginine vasopressin, and ACTH, by binding mostly on the mineralocorticoid but also on the glucocorticoid receptors located in various brain regions and in the anterior pituitary [10]. During the traumatic experience in PTSD, SNS interacts with HPA axis, and activation of the HPA is associated with the rapid release of noradrenaline, adrenaline, and dopamine, and cortisol binds also to glucocorticoid receptors [17, 18] that results in a failure of restoration to normal activity and in the failure of the feedback mechanism. This fine balance is altered and the HPA is dysregulated [17, 18], resulting in the abnormal and overstated reactions to the usual and non-frightening cues and stimuli, leading to the exaggerated fear reactions to stressors, fear potentiation and fear conditioning, consolidation of the traumatic memories, and inability of the fear extinction, a normal mechanism of fear inhibition [11, 15, 16, 19]. The HPA axis disruption is related to elevated CRH which might be responsible for the altered balance of glucocorticoids and CRH interactions, leading to the disrupted responses to traumatic and/or fear stimuli, or stressors and different symptoms in PTSD [20]. There are inconsistent data regarding plasma cortisol levels in PTSD [15], since the results differ according to the different body fluid sampled (e.g., plasma or saliva or urine), different time of the day when cortisol was measured, and various other factors that affect cortisol concentration such as sex, age, present therapy, or the time that period between sampling and exposure to a traumatic event [15, 21, 22]. There were no differences between PTSD patients and controls in cortisol levels in plasma, saliva, serum or urine, and no differences based on tissue type, but lower cortisol was detected in PTSD compared to trauma nonexposed controls [23]. Similarly, cortisol concentration did not differ in plasma, saliva, and urine in subjects with PTSD, trauma exposed controls and trauma nonexposed controls [24]. Regarding PTSD comorbid with major depression, morning cortisol levels and daily output cortisol levels were lower in PTSD and in subjects with PTSD and comorbid depression

compared to trauma unexposed controls, while evening cortisol level was reduced in PTSD and trauma exposed controls vs. trauma unexposed controls, but increased in PTSD comorbid with depression compared to trauma unexposed controls [25]. More uniform results are collected for saliva cortisol which is generally lower in PTSD subjects compared to controls [15, 22]. However, the level of ACTH did not differ between subjects with PTSD, trauma exposed controls and healthy subjects [26, 27]. The data about CRH in cerebrospinal fluid (CSF) are scarce and show elevated CRH release in PTSD [28, 29].

The HPA axis reactivity is associated with the activation of the immune system [30] and increased release of the pro-inflammatory cytokines [11, 31]. Chronic low-grade inflammation is present in PTSD [4–9]. Glucocorticoid receptors mostly inhibit and regulate proinflammatory cytokines. Among the immune markers, increased concentration of the acute phase reactant, C-reactive protein (CRP), was found in PTSD [4], and was related to the re-experiencing and avoiding symptoms [4, 32], the PTSD severity [2, 32], and a chronic form of PTSD [33]. Other pro-inflammatory markers associated with exposure to trauma are increased interleukin (IL)-1β and IL-6 and tumor necrosis factor (TNF)-α [34]. Most data suggest increased concentrations of IL-1β, IL-6, TNF-α, and interferon gamma (IFN-γ) in PTSD [35]; but the literature findings regarding these increased proinflammatory immune markers in PTSD are inconsistent, due to the variations in sample sizes, differences in the ethnicities involved, possible effects of therapy, presence of infection, and comorbidities with different mental and somatic conditions and in the comparison groups [11]. Therefore, both increased but also unchanged or reduced levels of interleukin IL-2, IL-6, IL-1β, CRP, TNF-α, and IFN-γ were detected in PTSD compared to controls, while anti-inflammatory markers IL-4, IL-8 and IL-10 are reduced in PTSD [11]. Recent meta-analysis suggested that increased levels of serum proinflammatory cytokines IL-1β, IL-6, and TNF-α might be used as potential markers of PTSD; however, serum IL-6 level is affected by the trauma subtype [3]. Inflammatory processes affect prefrontal cortex, amygdala, and hippocampus, regions altered in PTSD and associated with cognitive functions such as emotions, executive control, responses to fear, and retrieval of the fear- and trauma-induced memories. In these regions, but also in the HPA axis, increased IL-6 concentration might disturb the connection between proinflammatory cytokines and glucocorticoid receptors and their interaction [3].

There is a bidirectional interaction between inflammation and cognition, since inflammation disrupts cognition [36]; namely inflammation affects neuronal pathways involved in the regulation and response to fear, recall, and fear extinction and moderates cognitive processes [37], all processes disturbed in PTSD. In addition, there is also a bidirectional link between PTSD and cognition, since cognitive decline is a major symptom in PTSD [38], while cognitive deterioration might represent a risk factor for development, progression, or severity of PTSD [37]. However, the relationship between HPA axis and inflammation, i.e., inflammatory cytokines, is complex since both glucocorticoid receptors and cytokines modulate the HPA-immune axis via multiple feedback mechanisms achieved on different levels [39]. PTSD is associated with increased glucocorticoid receptor sensitivity

and elevated inflammation [39]. Glucocorticoids suppress proinflammatory cytokines and show anti-inflammatory effects at higher levels through increased glucocorticoid reactivity, while their proinflammatory responses are achieved at the basal glucocorticoid levels [39]. In addition, proinflammatory markers, TNF-α and IL-6, and high-sensitivity CRP are significantly associated with decreased cortisol levels after dexamethasone suppression test (DST) and post-DST cortisol decline but also with a promoter methylation of human glucocorticoid receptor gene exon 1F (*NR3C1-1F*) [39].

The mathematical modeling [39] of the HPA axis and the immune system revealed that elevated glucocorticoid sensitivity might lead to a higher inflammation. These results suggest that various strategies aimed to restore glucocorticoid sensitivity might be beneficial for PTSD since they might normalize inflammation [39].

## 11.3 PTSD, Inflammation, and Cardiovascular Disease

PTSD is frequently associated with cardiovascular disease (CVD), and it was recognized as early as in the Dutch World War II Resistance veterans [40] and confirmed today [41, 42]. A growing body of evidence has demonstrated the complex and multifactorial link between PTSD symptoms and poor somatic health, including CVD and coronary heart disease [41]. In a nationwide Swedish population-based and sibling-controlled follow-up study, people with stress related disorders, including PTSD, were at elevated risk of multiple CVD types, such as ischemic heart disease, cerebrovascular disease, heart failure, emboly, thrombosis, and fatal cardiovascular events [42]. The incidence of coronary heart disease was more than double in twins who had PTSD than in those without PTSD [43]. While traumatic stress induces numerous physiological changes, PTSD symptoms are those which are associated with CVD pathology. Middle-aged American war veterans with PTSD had increased risk for myocardial infarction, peripheral vascular disease, and congestive heart failure compared to veterans without PTSD [44]. The presence of PTSD was also linked to CVD risk factors: PTSD was independently associated with a worse endothelial function [45]. Among service members, participants who screened positive for PTSD had higher odds for hypertension [46]. PTSD is increasingly recognized as a systemic disorder [3]. For example, it is characterized by a higher nonpsychiatric healthcare utilization than the general population [47]. The link between CVD and PTSD is complex and includes common biological underpinning, shown in the increased prevalence of metabolic syndrome (MetS), hypertension, elevated pro-inflammatory cytokine and homocysteine levels, psychological mechanisms such as neuroticism and trait impulsivity/hostility, type-D (distressed) personality, and behavioral factors including unhealthy lifestyles, high smoking rates, and severe substances abuse [48].

Inflammation is supposed to have a critical role in the onset, progression, and manifestation of CVD, given that atherosclerosis is driven by a chronic inflammation [49]. Monocytes play a key role in the development of atherosclerotic plaques. They operate either directly, such as antigen presentation and cytokine secretion or

through their differentiation into macrophages or foam cells which secrete pro-inflammatory cytokines and chemokines [50]. Increased proinflammatory mediators, such as TNF-α, IL-1, and IL-6, along with CRP, contribute to the increased CVD risks via platelet activation and endothelial dysfunction and development and acceleration of atherosclerosis [48, 49]. Research on the long-term effects of psychological stress has focused on the immune system. Subclinical, low-grade inflammation may be a psychobiological link between PTSD symptoms and CVD.

Preclinical data provided important evidence on the devastating effects of stress on the heart. In a psychosocial stress model, stressed rats subjected initially to ischemia and after that to reperfusion had larger heart infarct sizes than non-stressed rats [51]. While trauma is quite essential for the PTSD onset, there is evidence that PTSD symptoms rather than trauma itself may induce heart tissue damage. In a predator stress model, rats that developed PTSD experienced histomorphological signs of metabolic and hypoxic injury in cardiomyocytes and impaired contractility, in contrast to PTST-resistant rats, who had no signs of cardiac injury [52]. Increased inflammation may also be triggered by PTSD symptoms, given that among the animals exposed to the same stress, those who developed PTSD-like symptoms had experienced signs of immunological dysfunction. Rats who presented with PTSD-like symptoms after predator stress had higher IL-6 and lower IL-4 in myocardium and plasma, compared to both control and PTSD-resistant rats [52]. In addition, TNF-α, IL-1β, and IL-6 were increased in the hippocampus of rats exposed to the single-prolonged stress compared to normal rats [53], and rats subjected to predator scent stress displayed higher TNF-α, IL-6, and IL-1β and several hypertensive component mRNA expressions in amygdala, but also higher plasma angiotensin II levels, than non-stressed rats [54].

Clinical data confirm that patients with PTSD had a trend for higher CSF IL-6 levels than trauma-exposed individuals without PTSD [3]. Among combat-exposed veterans, those with PTSD had higher cortisol and ACTH suppression, IL-6, TNF-α, and high sensitivity CRP and lower glucocorticoid receptor promoter methylation than participants without PTSD [39]. These studies suggest that traumatic stress, and particularly PTSD symptoms, is associated with the excess inflammatory response eventually leading to atherosclerosis.

Inflammatory changes might be the result of biological alterations related to PTSD symptoms. Multiple systems affect activity of the immune system, including HPA axis, sympathetic system, and sex hormones. This complex cascade of events begins with dysregulated amygdala activity. Amygdala is considered a starting point for these effects which transform negative emotional states to physiological effects. Stress increases microglial activity in the amygdala [55]. Amygdala activity increases during recollection of traumatic events, which induces HPA activation and the cascade of physiological responses to acute stress and later PTSD [56]. SNS predominance is well-determined finding in PTSD patients [57], together with the lower parasympathetic activity [58]. Preclinical studies demonstrated increased activity of both central and peripheral renin-angiotensin systems after experimental stress [55]. Patients with PTSD had lower morning and 24-h cortisol concentrations compared to healthy subjects [59]. Increased glucocorticoid receptor sensitivity, a

well-known finding in PTSD, may contribute to the increased inflammation due to its relationship with higher proinflammatory cytokine production [39]. While inflammatory cytokines normally upregulate cortisol secretion, in PTSD patients the glucocorticoid negative feedback prevails over cytokine-mediated positive feedback, resulting in insufficient cortisol rise and increased inflammatory response. A bidirectional relationship exists between the neuroendocrine and immune systems. Cortisol via glucocorticoid receptors inhibits inflammation, such as the production of TNF-α, IL-1, and IL-6 [60], while a decreased HPA axis function in PTSD results in the reduced ability of cortisol to inhibit inflammatory processes, leading to the increased release of pro-inflammatory cytokines and overactivity of the SNS. Patients with PTSD have increased heart rate, both at baseline and during stress [58]. Effects on blood pressure may, in turn, be more pronounced during stress. While the effects of PTSD symptom severity on blood pressure are not robust [61], blood pressure reactivity may be more important indicator of CVD risks. Namely, although rats exposed to predator scent stress had no changes in basal blood pressure and heart rate, rats had greater hypertensive response to a slow-pressor dose of angiotensin II challenge than non-stresses animals [55]. Importantly, this stress-related response was prevented by the TNF-α synthesis inhibitor pentoxifylline [55], suggesting an interplay between increased sympathetic activity and inflammation. Noradrenaline-dependent adrenergic stimulation results in nuclear factor-κappa B (NF-κB) activation in peripheral blood mononuclear cells that was reduced by α1- and β-adrenergic receptor inhibitors [62]. The release of proinflammatory cytokines from the activated mononuclear cells has numerous effects, such as decrease in serotonin availability and brain-derived neurotrophic factor (BDNF) levels [63], and the production of reactive oxygen species which may lead to heart damage [52]. General autonomic system dysregulation [58, 64] further drives immune dysfunction, that compromises the structural integrity of cardiac tissue [65]. Therefore, increased amygdala activity, which is a hallmark of PTSD, may contribute to the inflammation. In support, amygdala activity was linked to a systemic inflammation in the cohort consisting of PTSD subjects and healthy group, suggesting a presence of brain-systemic inflammation [12]. The activation of the brain renin-angiotensin system (RAS) or immune system can independently or synergistically lead to hypertension [55]. Preclinical study reported that stress induced the activation of the microglia in the rat hippocampus, accompanied by the increase in the hippocampal IL-1β, TNF-α, and IL-6 expression and decrease in anti-inflammatory IL-10 levels [53]. This link may be influenced by different factors, such as sex or the severity of PTSD symptoms. Namely, high circulating estrogen levels stimulate the HPA axis while inhibiting inflammation and the sympathetic activity [64].

Higher levels of systemic inflammation, presented as a combined inflammatory score, were found in patients with severe compared to those with moderate PTSD symptoms [66]. In addition, different stress reactivity was reported in patients with more severe symptoms, given that participants with severe PTSD symptoms had greater heart rate variability reduction than those with moderate symptoms [66]. CCF IL-6 levels also correlated positively with PTSD severity scores and independently contributed to PTSD severity [3]. These findings suggest that patients

with higher levels of psychopathology already have higher CVD risks, which further increase if they are exposed to stressful situations [66]. PTSD severity was also correlated with IL-6 levels [35]. In patients with recent myocardial infarction, higher PTSD symptom severity was associated with an enhanced inflammatory response of IL-6 to experimentally induced stress [67]. In addition, a longitudinal study found that persistently severe PTSD course was associated with a higher total white blood cell count [68]. The duration of PTSD was positively correlated with IL-1β levels [35]. These data collectively suggest a dose-response relationship between PTSD severity [35, 64, 67–69] or persistence of symptoms [35, 68] and the intensity of inflammation.

Another link between PTSD and inflammation may include poor health behavior. Namely, PTSD symptom severity was associated with eating poorer quality foods, mediated by emotion suppression, as an attempt to reduce the emotional burden [70]. In turn, poor diet may be related to obesity and inflammation. Being obese was associated with higher levels of serum TNF-α, IL-1β, and IL-6, all produced by macrophages from the adipose tissue [71]. While patients with PTSD were 31% more likely to be obese than participants in non-PTSD groups, of particular concern was the association between PTSD and obesity in individuals 20–30 years old [72]. Strikingly, in the same meta-analysis, the likelihood of current smoking in males with PTSD was highest in respondents in the same age group [72]. In agreement, stress-related disorders were strongly associated with an early onset CVD [42]. In Croatian war veterans, the majority of participants with PTSD who had no CVD were overweight and had total cholesterol and triglycerides slightly above reference range [73]. On the other hand, about one third of patients treated for PTSD, with the mean age of 55, have already been diagnosed with CVD [74]. Participants with PTSD were more frequently smokers [75] and had higher prevalence of tobacco dependence than veterans without PTSD [44]. Of note, twins with PTSD were also more frequently smokers, than twins without PTSD [76]. Heavy smoking may be associated with a more severe PTSD pathology [74]. There is also evidence that immunological dysfunction may contribute to a poor treatment outcome. Higher acute increase of plasma IL-6 levels after psychosocial stress prior to combined trauma-focused therapy was associated with a negative therapy outcome in PTSD, especially regarding depressive symptoms [77]. No differences in vascular or systemic inflammation, as assessed by fluorodeoxyglucose (FDG)-positron emission tomography (PET) imaging, were reported between PTSD and control subjects [12]. However, these data should be interpreted in the context of similar TNFα, IL-1β, and IL-6 levels, as well as lipids, glucose, blood pressure, BMI, and smoking prevalence across groups but also in a small number of patients and in the young (34 years) patient age [12]. In addition, older veterans had higher serum CRP levels than veterans in non-PTSD group, but after controlling for BMI and triglycerides, the significance of this association disappeared [78].

## 11.4 Cytokines in PTSD

Cytokines are glycoproteins with the major role in the initiation, amplification, mediation, and regulation of adaptive immunity [79]. They can act as proinflammatory factors (IL-1, IL-6, IL-12, TNF-α, and INF-γ), generating an inflammation in the host defense and disease, or they can have anti-inflammatory effect (IL-6, IL-10, and transforming growth factor beta (TGF-β)), by attenuating the inflammation and inducing repair [79]. At the periphery, cytokines are produced by B cells, T cells, macrophages, mast cells, neutrophils, basophils, and eosinophils [79], and in the central nervous system (CNS), they are produced by neurons, astrocytes, and microglia [80]. In CNS, cytokines can participate in physiological functions, such as neurite outgrowth, neurogenesis, neuronal survival, synaptic pruning, and regulation of synaptic plasticity [81], but also their overproduction and exaggerated release can be associated with neuronal dysfunctions related to neuropsychiatric disorders [82]. Since cytokines can cross blood-brain barrier, both inflammation in CNS and in the periphery can contribute to neuroinflammation [5]. In PTSD, the stress-mediated activation of HPA axis leads to elevated secretion of CRH which stimulates SNS to produce catecholamines, including noradrenaline which is associated with PTSD symptoms, such as hyperarousal. The complex interaction between the autonomic nervous system and the immune system is then manifested by the production of proinflammatory cytokines, such as IL-1 and IL-6, which is stimulated by an increased production of norepinephrine via NF-κB-dependent and other mechanisms [83].

A lot of studies have shown that individuals with PTSD exhibit significantly elevated blood levels of cytokines when compared to healthy control subjects [7, 8, 84, 85]. However, findings supporting the proinflammatory activity in PTSD are often inconsistent. The source of inconsistency can be of pure technical nature, such as different immunological assay methods or sampling procedures, or they can be related to gender, the type of included controls (exposed or non-exposed to trauma), or comorbid disorders and early-life adversities in PTSD patients. Although studies comparing the immunological factors between trauma-exposed individuals who developed PTSD and those who did not develop PTSD excluded this possibility [7], there are some indications that the trauma exposure itself can be the cause of the increased levels of proinflammatory markers [34, 85]. Namely, the levels of IL-6 and IL-10 were showed to be increased when PTSD patients were compared with no-trauma exposed controls, but they were not increased in the PTSD group when only trauma-exposed controls were used [85]. On the other hand, when PTSD patients were compared only to the control individuals not exposed to trauma, their levels of IL-1β were similar [85]. Compared to men, women are at higher risk of developing PTSD after trauma exposure [86], and it seems that pro-inflammatory cytokines are significant mediators in this relationship. A recent study [87] reported that, compared to women, men have higher total pro-inflammatory cytokine score (estimated from IL-6, IL-1β, TNF-α, and IFN-γ concentration), and it was associated with higher estradiol levels and lower risk of non-remitting PTSD development.

Beside gender, comorbid major depressive disorder (MDD) [88] and a history of childhood maltreatment [89] were showed to be associated with increased inflammation, which makes these conditions as confounding variables in the association between PTSD and inflammation. For example, a meta-analysis [85] indicated that, when comparing PTSD patients with healthy control individuals, IL-8 levels were elevated in PTSD patients only when MDD comorbidity was excluded.

It was also noticed that PTSD is prevalent in individuals who sustain traumatic brain injury (TBI) [90], but since this is not universal, one should question the source of this kind of variable vulnerability. The findings of a recent study [91] suggest that PTSD in military personnel and veterans with multiple TBIs is associated with chronic inflammation, specifically with chronically elevated levels of IL-6 in those individuals.

## 11.5 Oxidative Stress in PTSD

Oxidative stress is a molecular process underlying many chronic diseases and is closely related to the inflammatory process. It is caused by an excessive production and accumulation of reactive oxygen species (ROS) which exceeds the antioxidant capacity of a biological system, resulting in oxidative damage of cellular components and tissue.

There are different approaches to measure the level of oxidative stress [92]. One is to directly measure the cellular levels of ROS by specific fluorogenic probes. However, there are several ways to indirectly measure oxidative stress levels based on detecting the products of damaged biomolecules. It is possible to use protein carbonyl content as a marker of protein oxidation [92]. The level of lipid peroxidation is usually measured through malondialdehyde (MDA) formation or by identifying other lipid peroxidation products, such as 8-iso-prostaglandin F2α, 4-hydroxy-2-nonenal (4-HNE), conjugated dienes, and lipid hydroperoxides [92]. DNA damage caused by oxidative stress is quantified by measuring the level of 8-hydroxy-2′-deoxyguanosine (8-OHdG) which is generated due to hydroxylation of the deoxyguanosine residues [92]. It is also possible to measure the level of thymidine glycol or to evaluate DNA damage through single- or double-stranded breaks within the DNA [92]. Another approach for determining the extent of oxidative stress is to assess the antioxidant status of the cells by determining the activity of antioxidant enzymes that regulate ROS levels (superoxide dismutase (SOD), glutathione peroxidase (GPx), catalase) or by measuring the level of nonenzymatic antioxidants (glutathione, vitamin A, vitamin C, vitamin E). However, the total antioxidant status in clinical samples can also be determined with the help of different methods and assays [92].

Persistent exposure to psychologic stress, with emphasis on stressful events in childhood and adolescence, was associated with disrupted oxidant-antioxidant balance within the brain tissue and with the development of different psychiatric disorders [93]. Literature search revealed a modest number of studies that have focused on exploring the association between witnessing and/or experiencing

traumatic event and oxidative stress levels in humans that can be measured in a variety of ways, most commonly through specific markers of oxidative stress. Most of the studies that have focused on the involvement of oxidative stress in PTSD suggest its implication in the pathogenesis of PTSD.

Impaired antioxidant defense system, characterized by reduced levels of SOD and GPx, was detected in red blood cells of individuals diagnosed with PTSD, suggesting dysfunctional response to oxidative stress [94]. However, Tezcan and colleagues [95] reported no significant differences in SOD, GPx, and catalase activity between PTSD subjects and healthy individuals, but they detected a positive correlation between the PTSD symptom severity and the activity of SOD and GPx. The authors also reported a possible positive correlation between PTSD symptom severity and MDA levels [95]. The MDA, as a final product of lipid peroxidation in the cells, was found to be elevated in combat-related PTSD [96] and in civilians who developed PTSD after surviving earthquake [97]. Mentioned studies suggest that increased lipid peroxidation and decreased antioxidant enzyme function could be associated with PTSD pathophysiology [96, 97]. In our recent study, we have reported elevated levels of 4-HNE in patients with combat-related PTSD [98] which suggests that oxidative stress and subsequently altered lipid metabolism, reflected by an increase in 4-HNE levels, could be associated with pathophysiology of PTSD. However, there are also conflicting results that deny the association between lipid peroxidation (MDA) levels and PTSD, suggesting a possible compensatory mechanism and adaptation to stress in war veterans with PTSD, compared to veterans without PTSD diagnosis [94]. Michels and colleagues [99] found evidence of higher γ-aminobutyric acid and glutathione levels in anterior cingulate cortex and dorsolateral prefrontal cortex of subjects diagnosed with PTSD. Glutathione S-transferase mu1, an enzyme that participates in the metabolism of oxidative stress products, was also suggested as a potential marker for predicting PTSD development in US Marines [100, 101]. Increased levels of glycerophospholipids, phosphatidylethanolamine (PE; 18:1/0:0) and phosphatidylcholine (PC; 18:1/0:0) were detected in Croatian war veterans with combat-related PTSD [102]. The abovementioned lipid species are involved in the inflammation process and associated with membrane breakdown, oxidative stress, mitochondrial dysfunction, and neurotoxicity [102].

No relationship between chronic PTSD diagnosis and different markers of oxidative stress damage was reported in soldiers who participated in the Croatian war, in the period between 1991 and 1994 [103]. A similar conclusion was reached by the authors who investigated the association between oxidative stress levels and the development of PTSD after experiencing a sexual trauma [104]. However, their results indicated the correlation of lower levels of cortisol and 8-OHdG with the amount of time that has passed since the exposure to the trauma [104]. Urinary levels of 8-OHdG, along with serum thromboxane B2 and serum urates, were also investigated as potential biomarkers of oxidative stress in war veterans with PTSD, but no significant association was detected [103].

The relationship between PTSD diagnosis and oxidative stress is not yet clarified. One of the possible explanations can be found in abnormal functioning of HPA axis which we have long known to play an important role in the PTSD pathophysiology

[14]. However, the relationship between PTSD and oxidative stress could also be mediated by the sleep disturbances that are frequent in PTSD. There is evidence of increased MDA levels and reduced GPx activity in subjects with insomnia [105], while animal studies suggest that the lack of sleep can lead to hippocampal oxidative stress and memory deficits which can be reversed by the antioxidant agents, such as melatonin, N-tert-butyl-alpha-phenylnitrone, and vitamin E [106]. Sleep deprivation has also been linked to inflammation and proinflammatory markers, such as TNF-α, interleukins, and CRP [107, 108]. This leads to the conclusion that oxidative stress and inflammation may be the main processes behind accelerated aging, cognitive impairment, and neurodegeneration which are the long-term consequences of PTSD.

## 11.6 Chemokines in PTSD

Chemokines are small proteins, part of cytokine family involved in cell migration to the infection or injury sites [109] and initiation of inflammation [110]. In general, they are involved in the activation of inflammatory response, wound healing process, immune surveillance, localization, and migration of lymphocytes and leukocytes. According to the cysteine arrangement, chemokines might be divided into four subfamilies C, CC, CXC, and CX3C, while CXC and CC subfamilies are the most common chemokines in mammals [109, 111]. Alterations in the immune system, as well chemokines, have been reported for several neuropsychiatric disorders, including PTSD, which can be defined as psychoneuroimmunological disorder [111]. It is known that stress activates certain chemokines and receptors that might influence the HPA axis [112], which regulation has been altered in patients with PTSD. Hence, alterations in chemokines might lead to neuroendocrine and neurochemical alterations. Dysregulation of the HPA axis in combination with impaired immune system in patients with PTSD increase the risk for development of autoimmune, metabolic, and cardiovascular diseases [14]. Furthermore, chemokines can cross blood-brain barrier and cause certain alterations in brain areas associated with PTSD development and progression, including the amygdala, prefrontal cortex, hippocampus, and insula [113, 114]. Due to low-systemic chronic inflammation in patients with PTSD, several studies have found alterations in the immune system, including the role of chemokines [111]. For example, increased levels of chemokine CCL-5 have been reported in subjects with PTSD. CCL-5 is produced by macrophages, NK cells, platelets, T cells, and monocytes, while it is activated or inhibited by other cytokines [111].

In PTSD, it regulates arrival of monocytes and T-cells to the walls of brain vessels. Furthermore, in men and women with PTSD, CCL-5 levels were increased compared to healthy controls group, while the elevation was even higher for PTSD subjects with avoidant personality disorder [111]. The levels of another chemokine, SDF-1, were also reported as elevated in patients with PTSD compared with control group. Chemokine SDF-1 is produced by stromal cells in the bone marrow, and together with CXC receptor, it is highly expressed in the brain areas involved in the memory process and fear learning, such as amygdala, hippocampus, and

hypothalamus. Therefore, altered levels of chemokine SDF-1 might affect neuroendocrine regulation [111]. Moreover, the highest increase in the CCL-5 and SDF-1 chemokines has been found for women with PTSD and avoidant personality disorder. It is assumed that estrogens might have a role in the chemokine activation [111, 114]. Likewise, elevation has not only observed for chemokine levels, but also for their receptors, such as CCR-5 and CXCR-4. The levels of the receptor CCR-5 have been increased in both women and men with PTSD alone or in combination with avoidant personality disorder, compared with control subjects. Similar findings have been reported for the receptor CXCR-4 in women; however for men, no significant differences were found for the receptor CXCR-4 [111]. Therefore, chemokines CCL-5, SDF-1, and their receptors, CCR-5 and CXCR-4, might represent immunological but gender dependent biomarkers of chronic stress in PTSD. Furthermore, study published by Zhang and colleagues [115] reported altered levels of certain chemokines in subjects with PTSD compared to subjects without PTSD who were active in the military service. Elevation in the inflammatory state of PTSD subjects has been reported, which resulted in chemokine dysregulation. Four chemokines, CXCL-2, CCL-2, CCL-15, and CCL-22 were significantly increased, while CXCL-12 (SDF-1) and CCL-25 were significantly decreased in subjects with PTSD, compared with subjects without PTSD [115]. These chemokines might represent potential biomarkers for PTSD onset [115], while CCL-2 is increasing over the time in patients with PTSD [116]. Moreover, downregulation of CXL-6, CCL-13, and CCL-23, as well upregulation of CCL-20 might represent risk for PTSD, while dysregulation of the chemokines CXCL-11, CCL-13, CCL-23, and CCL-25 was associated with PTSD severity. The chemokine CCL-13 binds to the receptors on T-lymphocytes, basophils, eosinophils, and monocytes, while CCL-23 and CCL-25 bind to the specific receptors on monocytes, lymphocytes, or leukocytes in the peripheral blood [115]. Likewise, chemokine CX3CL-1 might be associated with reduced PTSD risk. Its lower levels have been reported in subjects with PTSD, in comparison with subjects without PTSD [115]. CX3CL-1 is a large chemokine with important role in migration adhesion, learning, neurotransmission, and synaptic plasticity. It is assumed that alterations in the chemokines and interactions with other cytokines might be involved in alterations of certain brain regions that regulate fear conditioning and memory processes, such as the prefrontal cortex, insula, and amygdala [114]. Alterations in these brain areas are associated with PTSD symptomatology. Activation of the astrocyte's proliferation leads to chemokine release and production of ROS, which might interrupt synthesis of serotonin. Decreased levels of serotonin might be associated with typical PTSD symptoms, including re-experiencing and hyperarousal. Hence, according to the published results, it is assumed that elevation of inflammatory reaction, as well altered levels of chemokines and others mediators of immune system are associated with PTSD development and progression [111]. However, it should be considered that differences in the chemokine levels might be due to gender differences, sample type, age, and detection methods [114]. For example, certain sex-specific variations have been found in the genes for chemokines, while different endocrine regulation might also lead to different chemokine levels between women and men. Regarding

the age, significant differences in chemokines were found in older PTSD subjects, probably due to imbalance in immune system. Moreover, reported by Pan and colleagues [114], certain detection methods, such as ELISA, are much more stable and sensitive for detection of chemokine alterations, while plasma samples are more suitable for chemokine detection in comparison to serum samples.

## 11.7 CRP and PTSD

Immunological disruptions, especially in innate immune pathways, have been linked to increased risk of PTSD and more severe clinical manifestation of PTSD symptoms [117]. One of the most investigated and validated markers of the innate immune signaling pathways is CRP which is synthesized primarily in the liver during acute-phase inflammation, and its concentration drastically elevates within 2 h after inflammatory trigger [118, 119]. CRP exhibits its proinflammatory properties through stimulation of the complement system, activation of macrophage phagocytosis, and reduction of the anti-inflammatory IL-10 levels [117, 120]. Elevated concentration of peripheral CRP has been associated with increased risk of CVD, metabolic syndrome, diabetes, hypertension, and other chronic diseases [121].

Although some studies showed lower levels of CRP in PTSD [122], a majority of studies reported increased levels of plasma CRP in both civilian and military subjects with PTSD [2, 123–126]. Moreover, patients with PTSD were two times more likely to have clinically significant increase in CRP levels (>3.0 mg/L), associated with cardiovascular risk and metabolic syndrome, which are both common comorbidities in PTSD patients [125]. Recent studies have shown increased CRP in stroke lesions [127], senile plaques in patients who suffered from Alzheimer's disease [128–130], and CSF and plasma of patients with MDD [131, 132]. The role of CRP in immunological and pathological states was mostly associated with peripheral signaling pathways, while conclusive mechanisms by which it could affect the CNS and psychiatric disorders remain unclear. It has been suggested that CRP could also been produced by endothelial cells that form the blood-brain barrier (BBB) [133] or that increased BBB permeability, which has been reported in states of high inflammation and TBI could cause the crossing of peripheral CRP to CSF and brain [134–136]. That way, not only CRP, but other pro-inflammatory cytokines and immune cells could enter the brain and contribute to the neuroinflammation and PTSD risk and symptoms severity [117].

There is growing evidence of the association of PTSD and CRP levels; however, it is unclear whether the increased pre-trauma CRP is a risk factor contributing to the development of PTSD or it is a clinical marker of already developed PTSD symptoms. Increased CRP was reported in male Marines before combat deployment who subsequently developed PTSD; however, it did not correlate with PTSD severity [137]. On the other hand, pre-trauma levels of CRP were not significantly predictive for PTSD development in civilian women [138]. These differences are not surprising since alterations in innate immune system response, including CRP levels, have already been documented between men and women, as well as between

different ethnicities [117, 139, 140]. Elevated CRP was associated with several symptom domains, mostly with avoidance, re-experiencing [4, 141, 142], and depression [32] but also with impaired inhibition of fear-potentiated startle in women, which is also related to PTSD symptoms, specifically with hyperarousal [2, 143]. The association of increased CRP and heightened fear response and fear of a terrorist event was more noticeable in women than in men, while the depressive symptoms after terrorist-induced trauma were more correlated with CRP in male civilian population compared to female subjects [144].

Although there is possibility that traumas involving physical injury such as war experience, car accidents, and physical violence, would activate stronger immune response than psychological trauma [145], the differences in CRP levels depending on the types of traumas have not been directly studied. However, the history of childhood trauma is associated with higher inflammation and CRP levels in adulthood [146–148] and also with higher risk of psychiatric disorders, including PTSD [11, 149]. Other environmental factors such as socioeconomic status and social support could also mediate the CRP association with PTSD development. Low socioeconomic status was associated with worse clinical manifestation of PTSD [150] and with higher CRP levels [151, 152]. Additionally, low socioeconomic status has been associated with lower DNA methylation of several proinflammatory genes, which could lead to higher expression of proinflammatory factors and exacerbated inflammatory response [153]. Social support could influence the trauma perception, processing, and management and in that manner might alleviate the symptoms or reduce the risk for development of PTSD after traumatic event [154, 155]. Patients who received high social support also had lower levels of CRP [156, 157].

The estimated heritability of CRP is around 35–40% [158], and recent studies have shown the association of rs1130864, SNP within CRP gene, and CRP levels, as well as with PTSD symptoms, especially increased hyperarousal, vulnerability to hypervigilance, increased fear-potentiated startle [2], and major depression [159]. Additionally, SNP rs3091244, which influences CRP promotor activity and is in strong linkage disequilibrium (LD) with rs1130864, has been associated with CRP levels in healthy individuals [160] and subjects with PTSD [2], while rs1205 and rs2794520, previously associated with cardiometabolic conditions [161], significantly interacted with PTSD to influence CRP levels [162]. Decreased methylation at cg10636246, located near the transcription start site of absent in melanoma 2 gene (AIM2), which plays a role in activating the innate immune response [158, 163], was associated with both increased expression of AIM2 and elevated CRP levels, as well as with PTSD severity [162].

These results suggest that association of PTSD and CRP is bidirectional and is possibly mediated by genetic variations in the CRP gene, but also with other factors such as gender, type of trauma, history of maltreatment in young age, socioeconomic status, dietary habits, and social support, that could influence the CRP levels and its relationship in PTSD pathology partially on epigenetic level [117, 162, 164]. Increased inflammation, which can be reflected in increased peripheral CRP, could reflect the symptoms that cross between different neuropsychiatric disorders;

however, the exact mechanism by which it could affect the trauma management in the CNS is still unknown. More in vivo and in vitro studies are necessary to understand the role of CRP and other immune signaling factors in PTSD development and severity [117].

## 11.8 Stress-Related Regulation of the Kynurenine Pathway

The kynurenine pathway represents an important link between stress, CNS, and neuroendocrine and immune systems, as well as altered behavior [165–168]. This pathway regulates many important biological systems including oxidative stress, energy metabolism, immune function, gut-microbiota actions, and neurotransmitter systems [169]. Moreover, the kynurenine pathway is activated by acute and chronic stress and immune responses, and resulting neuroactive and immunomodulatory kynurenines may be involved in the etiology of a wide range of illnesses including immune diseases, cancer, neurodegenerative diseases, and psychiatric disorders [168, 170–175].

Kynurenine pathway is, in addition to protein synthesis and serotonin/melatonin production, the main metabolic pathway of the essential amino-acid tryptophan [166, 173, 176]. Approximately 95–99% of tryptophan is metabolized through the kynurenine pathway [174]. The enzymes, tryptophan 2,3-dioxygenase (TDO), and indoleamine 2,3-dioxygenase (IDO) [177], both metabolize tryptophan to kynurenines; however, TDOs are primarily distributed in the liver, whereas IDOs are found in the brain, blood, lung, spleen, and kidney [174, 178]. Kynurenine (KYN) is subsequently converted by the enzyme kynurenine 3-monooxygenase (KMO) into metabolites, which exert modulatory effects on glutamatergic neurotransmission [166] as well as various immune effects [179]. Specifically, in the so-called excitotoxic branch, neurotoxic kynurenine metabolites are generated: 3-hydroxy-kynurenine (3-HK) is metabolized into quinolinic acid (QUIN), subsequently to 3-hydroxy-anthranilic acid (3-HAA), and then to the end-point metabolite nicotinamide adenine dinucleotide (NAD+) [176, 180]. On the other hand, in the so-called neuroprotective branch, KYN is metabolized by the kynurenine aminotransferase (KAT) into kynurenic acid (KYNA), the NMDA, and alpha7 nicotinic acetylcholine receptor antagonist [166, 180, 181]. KYNA exerts neuroprotective, antioxidant, and immunomodulatory properties [182–184], by reducing extracellular release of glutamate [185, 186] and decreasing inflammation, oxidative imbalance, and mitochondrial dysfunction [183, 187] and might counteract the neurotoxicity mediated by 3-HK and QUIN [166]. The balance between KYNA and QUIN is suggested as an important parameter of the brain homeostasis [188], whereas its disruption underlies the pathogenesis of different CNS disorders, such as mood and anxiety disorders and certain stress-related diseases [189, 190].

Stress activates pro-inflammatory cytokines [191], such as TNF-α, IL-1β, IL-6, and IFN-γ, [192–194], and cortisol secretion via stimulation of the HPA axis, which activates IDOs and TDOs, respectively [195–197]. Specifically, during stress IDOs are activated by proinflammatory cytokines, whereas TDOs are activated by the

glucocorticoids [174], resulting in the shift in the tryptophan metabolism from the methoxyindole pathway to the kynurenine pathway [174]. As a result, the kynurenine formation is elevated and neurotoxic downstream kynurenine metabolites such as 3-hydroxy-kynurenine (3-HK) and quinolinic acid (QUIN) are increased [198], leading to the induction of free radicals and neuronal apoptosis [194]. QUIN, a N-methyl-D-aspartate (NMDA) receptor agonist, has been reported to increase glutamate levels and induce oxidative stress and ROS formation, mitochondrial dysfunction, and reduced respiratory capacity, resulting in neuronal excitotoxicity and apoptosis [181, 199–202]. Neurotoxicity is also caused by the oxidative properties of 3-HK and of the end-product metabolite NAD+ [203]. Several kynurenine metabolites have also neuroactive properties and are involved in regulation and modulation of neurotransmitter systems such as glutamatergic, GABAergic, nicotinic, serotonergic, and dopaminergic systems [169]. Therefore, they may influence neurotransmission and neuronal function [174] and contribute to the development of CNS disorders. In addition, stress and inflammatory states induced via kynurenine pathway activation may trigger serotonin deficiency, by depleting the tryptophan resources necessary for serotonin synthesis, resulting in mood and psychiatric symptoms [204–206].

Various human studies [173, 207, 208], as well as experiments in animal models [209], have shown that the kynurenine pathway is activated during acute and chronic stress. In addition, several clinical studies have found the increase in kynurenine in the peripheral circulation to be associated with CNS diseases and might serve as a reliable biomarker to highlight dysregulation in the kynurenine metabolic pathway [169, 210, 211]. In addition, pharmacological agents targeting specific kynurenine pathway enzymes have been investigated in animal models of CNS disorders, in order to offer novel therapeutic targets [212, 213]. Different types of stressors, such as acute predatory stress [214], physical restraint or immobilization [215, 216], foot shock [217], separation stress [218], or different types of chronic stress [219, 220], activate the kynurenine pathway, suggesting that this pathway represents a good candidate to mediate the effect of stress on brain neurotransmission, by generating a variety of metabolites acting as oxidants, antioxidants, neurotoxins, neuroprotectants and immunomodulators [175]. It has been shown that the immunomodulators facilitate the immune system resulting in a chronic low-grade inflammation, which is commonly observed in obesity, poor nutrition, after exposure to chemicals or allergens, as well as in various chronic disorders, such as cardiovascular, metabolic, immune, neurodegenerative, and psychiatric diseases [221], including PTSD [9].

Inflammation, and particularly systemic low-grade inflammation, has been associated with PTSD [9]. Elevated levels of proinflammatory cytokines IL-1, IL-6, IFN-γ, and TNF-α have been observed in the serum of patients with PTSD and correlated with the severity of the disease [3]. On the other hand, lower serum concentrations of anti-inflammatory cytokine IL-4 were also reported in subjects with PTSD [113]. Moreover, higher anti-inflammatory cytokine TGF-β was found as predicative indicator for the development of PTSD 1 month after accidents [222]. However, so far there are no published data on the alterations in the kynurenine pathway in PTSD [11], and there are no clinical studies regarding the

peripheral or CSF samples of kynurenines in PTSD patients. The kynurenine pathway metabolites are monitored in clinic as evidence of inflammatory responses contributing to the sleep deprivation and the formation of intrusive memories [113]. Nevertheless, in rodents, injections of TNF-α and IL-6 in the amygdala resulted in the glutamate toxicity, which is associated with impaired auditory fear conditioning [223, 224]. In addition, the altered levels of neurotransmitters, such as GABA [225] in PTSD and other fear- and anxiety disorders, might be linked to inflammation-induced glutamate excitotoxicity [226].

Therefore, although there is accumulating evidence for the kynurenine pathway activation by the stress and immune responses in various neuropsychiatric and neurodegenerative disorders, more research is needed, especially regarding its involvement in PTSD and its potential role as a target for drug discovery and development.

## 11.9 The Gut-Brain Axis in PTSD

The microorganisms composing gut bacteria, or gut microbiota, play an important role in maintaining health and influence the brain through complex bidirectional communication via immune, neurological, and endocrine pathways, known as the gut-brain axis. More than 1000 species of gut microbiota are located in the human intestine, with $10^{11}$ to $10^{12}$ bacteria per gram of stool. Research conducted on gut microbiota and their host brain function has revealed that gut microbiota affects stress and emotional responses, as well as psychosomatic disorders [227].

Experiments on germ-free mice have discovered that the gut microbiome is a new major player in the structure, development, and function of both the enteric and central nervous system [228], while experimental and clinical data suggest that intestinal bacteria mediate changes in brain function and behavior such as depression, anxiety, and cognition. Germ-free mice showed reduced anxiety-like behavior [229] and impaired working memory [230] compared to normal, conventionally raised mice. Therefore, behavioral traits of a more anxiety-like phenotype could be adoptively transferred to mice that showed a less anxious phenotype by colonization with their donor gut bacteria [231]. Furthermore, specific strains of beneficial bacteria such as *Lactobacillus rhamnosus* or *Bifidobacterium longum* can ameliorate anxiety- and depressive-like behaviors after their administration to mice [232, 233]. Modification of the gut microbiota causes these changes in behavior that are associated with changes in brain neurochemistry including changes in the BDNF and N-methyl-D-aspartate receptor expression, but this communication between intestinal bacteria and brain is highly complex and involves several metabolic, neural, and immune pathways [229, 231]. Gut microbiota is also characterized by a huge metabolic activity. They ferment and digest host-derived and dietary components (carbohydrates, proteins, and lipids) and convert them into various metabolites that can be beneficial or harmful for health [234]. Capsular polysaccharide A, a membrane component of *Bacteroides fragilis*, and membrane vesicles from the cell surface of *Bacteroides fragilis* of *Lactobacillus rhamnosus*

*JB-1* can have anti-inflammatory and neuronal effects that represent effects of parent bacteria [235]. Also, the production of systemic serotonin via the tryptophan/kynurenine pathway is dependent on the presence of gut bacteria, and consumption of a probiotic (*Bifidobacterium infantis*) changes this pathway and is associated with antidepressant effects in a rodent study [236]. Food antigens, possible pathogens, and symbiotic intestinal microbiota that the gastrointestinal tract is constantly confronted with present a risk factor for intestinal inflammation. The gastrointestinal tract is highly innervated by vagal fibers that connect the CNS with the intestinal immune system. Anti-inflammatory capacities of the vagus nerve, another important intermediary component in the gut-brain communication, are mediated through the HPA axis, the splenic sympathetic anti-inflammatory pathway, and the cholinergic anti-inflammatory pathway. This cholinergic anti-inflammatory pathway plays a crucial role in the intestinal immune response and homeostasis and presents an interesting target for the development of novel treatments for inflammatory diseases such as PTSD, related to the gut-immune system [237]. Further, investigations on gut microbiota in patients with MDD have reported increased levels of *Alistipes* and *Oscillibacter*, reduced levels of *Faecalibacterium* [238], and increased fecal levels in bacterial product isovaleric acid [239]. These findings suggest that anxious and depressive symptoms, common in PTSD, may be associated with composition and functionality alterations of the gut microbiota. Thus, the balance of communities of commensal bacteria is important in the regulation of the gut barrier function, immune, and nervous systems that in turn can affect brain function and behavior [236].

In the exploratory study conducted on 18 PTSD participants and 12 trauma-exposed control participants from South Africa, no significant differences in diversity of a microbial community or predicted functional capacity between these two groups were found [240]. However, in this study, random forest analysis has revealed three phyla, *Actinobacteria*, *Lentisphaerae*, and *Verrucomicrobia* that distinguish PTSD participants from trauma-exposed controls. Higher PTSD clinician administered PTSD scale (CAPS) scores were associated with a decreased total abundance of these three phyla [240]. The *Verrucomicrobia* phylum was represented by *Akkermansia muciniphila* which is thought to be anti-inflammatory in humans, induces T regulated (Treg) cells, and is reduced in many diseases or conditions associated with a failure of immunoregulation and/or increased inflammation [240]. The *Actinobacteria* was represented by *Collinsella* genus, and its decreased relative abundance has been reported in individuals with MDD [238]. Further, individual differences in the host immune response possibly play an important role in the vulnerability to PTSD after trauma exposure. Studies in rats showed that glucocorticoids decrease immunoglobulin A (IgA), which is responsible for inhibition of bacterial adherence to intestinal epithelial cells, and they increase bacterial adherence, as well as bacterial translocation to mesenteric lymph nodes [240]. Decreased frequency of Treg cells or altered Treg function may result in overactive host immune defenses, increased gut permeability, colitis, and exaggerated PTSD symptoms after trauma exposure [241]. According to this study, decreases in the relative abundances of *Actinobacteria*, *Lentisphaerae*, and

*Verrucomicrobia* with the prevalent human commensal *Akkermansia muciniphila* could contribute to decreased immunoregulation in PTSD [240].

Dietary interventions or the use of beneficial bacteria such as probiotics can potentially improve the composition and the function of the bacterial community in the gut [236]. Also, the administration of different species of *Lactobacillus* and *Bifidobacterium* was found to be associated with an improvement in mood, a decrease in anxiety, as well as a decrease in psychological distress, especially in individuals with low cortisol levels. Furthermore, the administration of probiotics from fermented milk products affects the activity of brain regions that are responsible for the central processing of emotions in women [236]. Bio-immunomodulatory probiotics, such as *Lactobacillus reuteri* DSM 17938, are of greater interest due to their potential to decrease stress-induced inflammatory responses, high accessibility, low costs, self-sustaining, and existing information about their previous safety and tolerability without serious side effects [242]. Their ability to induce the proliferation of Treg cells and to increase the production of anti-inflammatory cytokines, including IL-10 and TGF-β, makes them promising candidates for the treatment of PTSD symptoms accompanied by mild traumatic brain injury [242].

## 11.10 Potential Treatment

The most effective treatment in PTSD is trauma-focused psychotherapy, but unfortunately even 30–50% of patients do not benefit from it [243]. It was shown that while during therapy symptoms became less severe, cytokine levels were increasing until the anti-inflammatory therapy was applied [116]. A more recent study [77] investigated the predictive associations of acute stress-induced IL-6 reactivity before the onset of psychotherapy intervention and the therapy outcome after 8 weeks of treatment. They reported [77] an association of the high reactivity of IL-6 to psychosocial stress at the beginning of the therapy with a negative therapy outcome in PTSD, especially regarding depressive symptoms.

There is an urgent need to determine the effect of bacterial supplements and controlled changes in diet on psychological symptoms and cognitive functions in patients with PTSD. Dysregulated HPA axis and altered glucocorticoid signaling suggest that future studies should focus on the strategies aimed to restore glucocorticoid sensitivity to normalize inflammation, since glucocorticoid receptors inhibit and regulate proinflammatory cytokines [39].

Although the link between PTSD, inflammation, and CVD is established in majority of studies, there is paucity of data how potential interventions may impact the CVD and inflammation in this population. In preclinical study, both ACE or TNF-α inhibitors prevented proinflammatory and hypertensive response to stress [55], while suppressing sympathetic tone by clonidine or blocking β-adrenergic receptors by propranolol did not block myocardial hypersensitivity to ischemia during psychosocial stress [51]. Moreover, lycopene has suppressed the increase of IL-6, IL-1β, and TNF-α and decrease of BDNF after single prolonged stress in mice hippocampus and prefrontal cortex [244]. In veterans with PTSD, a

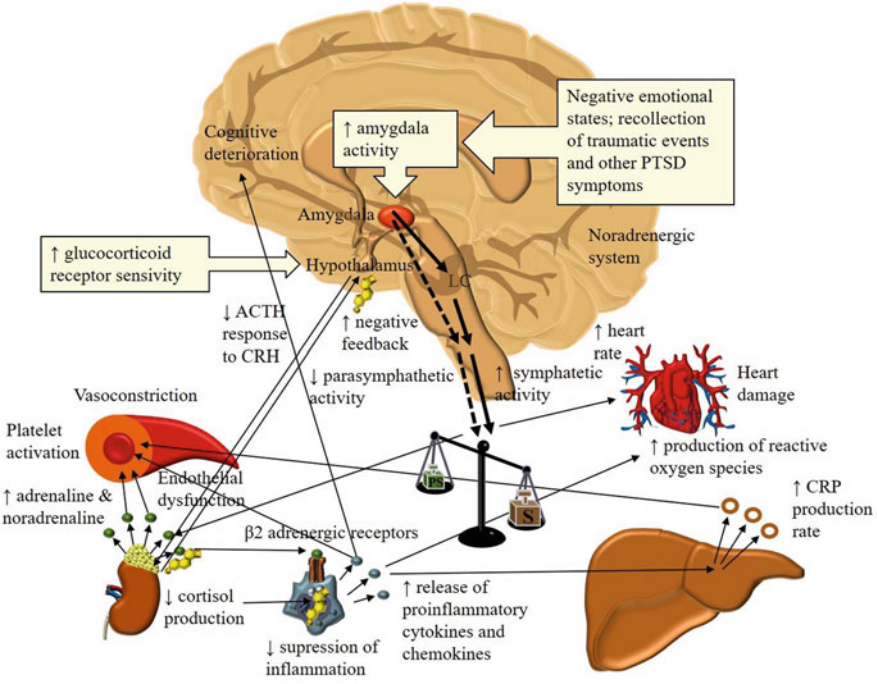


**Fig. 11.1** The complex link between PTSD symptoms and increased inflammation

combination of different treatments, including psychotherapy, increased serum TNF-α levels, despite improvements of clinical symptoms [245]. Likewise, serum IL-1β, chemokine MCP-1 and TNF-α levels in PTSD patients increased during 12-week multidisciplinary program, in contrast to other patients [116]. However, in another study including PTSD patients, serum IL-1β levels were decreased to normal levels after treatment with citalopram and sertraline but also after placebo [246]. There is also evidence that the modulation of autonomic response, such as vagal nerve stimulation, may improve the vagal tone and block the increase in mental-stress related cytokine increase [63].

PTSD is a chronic, difficult-to-treat disorder. Given its relationship with a range of somatic disorders such as CVD, and the role of low-grade inflammation, it remains to be determined how different treatments impact long-term CVD risks.

Since some individuals are resilient to trauma and do not develop PTSD after exposure to a traumatic experience, these differences might suggest that distinct pre-existing proinflammatory state might be responsible for the vulnerability or resilience to develop PTSD after traumatic exposure [3, 69], as shown in Fig. 11.1.

## 11.11 Conclusion

It is obvious that a great attention was given to the inflammatory pathology of PTSD, but there are still a lot of details in inflammatory mechanisms that remain to be investigated. Alterations in chemokines and interactions with other cytokines might be involved in alterations of certain brain regions that regulate fear conditioning and memory processes, such as the prefrontal cortex, insula, and amygdala. Changes in these brain areas are associated with PTSD symptomatology. However, there is a great potential in using altered inflammatory indicators as potential diagnostic and prognostic biomarkers of PTSD. They can also be used in searching for the novel anti-inflammatory treatment strategies in PTSD. The relationship between PTSD diagnosis and oxidative stress is not yet clarified. However, there is evidence linking chronic and repeated activation of the HPA axis with oxidative stress and inflammation. This leads to the conclusion that oxidative stress and inflammation may be the main processes behind the accelerated cellular aging and neurodegeneration, which are the long-term consequences of PTSD, and highlights the importance of future research that would focus on the novel therapeutic approach to PTSD, targeting both oxidative stress and inflammation. In addition, there is accumulating evidence for the significance of the kynurenine pathway activation by the stress and immune responses in various neuropsychiatric and neurodegenerative disorders. However, more research is needed, especially regarding its involvement in PTSD and its potential role as a target for drug discovery and development. In addition, the findings suggest that association of PTSD and CRP is bidirectional and is possibly mediated by the genetic variations in the CRP gene but also with other factors such as gender, type of trauma, history of maltreatment in young age, socioeconomic status, dietary habits, and social support, which could influence the CRP levels and its relationship in PTSD pathology on epigenetic level. Increased inflammation, which is reflected by the increased peripheral CRP levels, could reflect the symptoms that cross between different neuropsychiatric disorders; however, the exact mechanism by which it could affect the trauma management in CNS is still unknown. The gut microbiota maintains the balance and interaction of the immune, CNS, and endocrine pathways. Some bacteria might distinguish PTSD participants from trauma-exposed controls and might contribute to immune dysregulation in PTSD. Dietary interventions or the use of beneficial bacteria such as probiotics can potentially improve the composition and the function of the bacterial community in the gut, and therefore the effect(s) of bacterial supplements and controlled dietary changes on psychological symptoms and cognitive functions in patients with PTSD needs to be determined.

The dysregulated HPA axis, altered immune signaling, and disrupted homeostasis, as well as the association of the PTSD with the inflammation and disrupted cognition, oxidative stress markers, disrupted brain-gut axis, and CVD, support novel strategies, and new avenues for treatment of PTSD. These strategies should be aimed to attenuate inflammatory processes and consequently to reduce PTSD symptoms, but also to improve cognition and reduce cardio-metabolic disorders associated so frequently with PTSD.

## References


1. Britvić D, Antičević V, Kaliterna M, Lušić L, Beg A, Brajević-Gizdić I, Kudrić M, Stupalo Ž, Krolo V, Pivac N. Comorbidities with posttraumatic stress disorder (PTSD) among combat veterans: 15 years postwar analysis. Int J Clin Health Psychol. 2015;15:81–92.
2. Michopoulos V, Rothbaum AO, Jovanovic T, Almli LM, Bradley B, Rothbaum BO, Gillespie CF, Ressler KJ. Association of CRP genetic variation and CRP level with elevated PTSD symptoms and physiological responses in a civilian population with high levels of trauma. Am J Psychiatry. 2015;172:353–62.
3. Kim BK, Fonda JR, Hauger RL, Pinna G, Anderson GM, Valovski IT, Rasmusson AM. Composite contributions of cerebrospinal fluid GABAergic neurosteroids, neuropeptide Y and interleukin-6 to PTSD symptom severity in men with PTSD. Neurobiol Stress. 2020;12: 100220.
4. Canetti D, Russ E, Luborsky J, Gerhart JI, Hobfoll SE. Inflamed by the flames? The impact of terrorism and war on immunity. J Trauma Stress. 2014;27:345–52.
5. Hori H, Kim Y. Inflammation and post-traumatic stress disorder. Psychiatry Clin Neurosci. 2019;73:143–53.
6. Kibler JL, Tursich M, Ma M, Malcolm L, Greenbarg R. Metabolic, autonomic and immune markers for cardiovascular disease in posttraumatic stress disorder. World J Cardiol. 2014;6: 455–61.
7. Lindqvist D, Dhabhar FS, Mellon SH, Yehuda R, Grenon SM, Flory JD, Bierer LM, Abu-Amara D, Coy M, Makotkine I, Reus VI, Bersani FS, Marmar CR, Wolkowitz OM. Increased pro-inflammatory milieu in combat related PTSD - a new cohort replication study. Brain Behav Immun. 2017;59:260–4.
8. Lindqvist D, Mellon SH, Dhabhar FS, Yehuda R, Grenon SM, Flory JD, Bierer LM, Abu-Amara D, Coy M, Makotkine I, Reus VI, Aschbacher K, Bersani FS, Marmar CR, Wolkowitz OM. Increased circulating blood cell counts in combat-related PTSD: associations with inflammation and PTSD severity. Psychiatry Res. 2017;258:330–6.
9. Speer K, Upton D, Semple S, McKune A. Systemic low-grade inflammation in post-traumatic stress disorder: a systematic review. J Inflamm Res. 2018;11:111–21.
10. Gjerstad JK, Lightman SL, Spiga F. Role of glucocorticoid negative feedback in the regulation of HPA axis pulsatility. Stress. 2018;21:403–16.
11. Michopoulos V, Powers A, Gillespie CF, Ressler KJ, Jovanovic T. Inflammation in fear- and anxiety-based disorders: PTSD, GAD, and beyond. Neuropsychopharmacology. 2017;42: 254–70.
12. Toczek J, Hillmer AT, Han J, Liu C, Peters D, Emami H, Wu J, Esterlis I, Cosgrove KP, Sadeghi MM. FDG PET imaging of vascular inflammation in post-traumatic stress disorder: a pilot case-control study. J Nucl Cardiol. 2021;28:688–94.
13. Agorastos A, Pervanidou P, Chrousos GP, Baker DG. Developmental trajectories of early life stress and trauma: a narrative review on neurobiological aspects beyond stress system dysregulation. Front Psych. 2019;10:118.
14. Pivac N, Konjevod M, Sagud M, Uzun S, Kozumplik O. Neuroendocrine and immune biomarkers of posttraumatic stress disorder in combat veterans. In: Kumar U, editor. The Routledge international handbook of military psychology and mental health. 1st ed. Routledge, Abingdon, Oxon; New York, NY, Routledge, Taylor & Francis Group; 2020. p. 483–96.
15. VanDyke LM, Burton LE, Hamidovic A, Burge MR. Hormonal aspects of post-traumatic stress disorder. Endocr Diab Metab J. 2017;1:1–10.
16. Yehuda R, Hoge CW, McFarlane AC, Vermetten E, Lanius RA, Nievergelt CM, Hobfoll SE, Koenen KC, Neylan TC, Hyman SE. Post-traumatic stress disorder. Nat Rev Dis Primers. 2015;1:15057.

17. Daskalakis NP, McGill MA, Lehrner A, Yehuda R. Endocrine aspects of PTSD: hypothalamic-pituitary-adrenal (HPA) axis and beyond. In: Comprehensive guide to post-traumatic stress disorder. Springer; 2015. p. 245–60.
18. de Kloet CS, Vermetten E, Heijnen CJ, Geuze E, Lentjes EG, Westenberg HG. Enhanced cortisol suppression in response to dexamethasone administration in traumatized veterans with and without posttraumatic stress disorder. Psychoneuroendocrinology. 2007;32:215–26.
19. Osório C, Probert T, Jones E, Young AH, Robbins I. Adapting to stress: understanding the neurobiology of resilience. Behav Med. 2017;43:307–22.
20. Raglan GB, Schmidt LA, Schulkin J. The role of glucocorticoids and corticotropin-releasing hormone regulation on anxiety symptoms and response to treatment. Endocr Connect. 2017;6: R1–7.
21. van den Heuvel LL, Wright S, Suliman S, Stalder T, Kirschbaum C, Seedat S. Cortisol levels in different tissue samples in posttraumatic stress disorder patients versus controls: a systematic review and meta-analysis protocol. Syst Rev. 2019;8:7.
22. Pan X, Wang Z, Wu X, Wen SW, Liu A. Salivary cortisol in post-traumatic stress disorder: a systematic review and meta-analysis. BMC Psychiatry. 2018;18:324.
23. Meewisse ML, Reitsma JB, de Vries GJ, Gersons BP, Olff M. Cortisol and post-traumatic stress disorder in adults: systematic review and meta-analysis. Br J Psychiatry. 2007;191:387–92.
24. Klaassens ER, Giltay EJ, Cuijpers P, van Veen T, Zitman FG. Adulthood trauma and HPA-axis functioning in healthy subjects and PTSD patients: a meta-analysis. Psychoneuroendocrinology. 2012;37:317–31.
25. Morris MC, Compas BE, Garber J. Relations among posttraumatic stress disorder, comorbid major depression, and HPA function: a systematic review and meta-analysis. Clin Psychol Rev. 2012;32:301–15.
26. Muhtz C, Wester M, Yassouridis A, Wiedemann K, Kellner M. A combined dexamethasone/corticotropin-releasing hormone test in patients with chronic PTSD-first preliminary results. J Psychiatr Res. 2008;42:689–93.
27. Savic D, Knezevic G, Damjanovic S, Spiric Z, Matic G. The role of personality and traumatic events in cortisol levels-where does PTSD fit in? Psychoneuroendocrinology. 2012;37:937–47.
28. Baker DG, West SA, Nicholson WE, Ekhator NN, Kasckow JW, Hill KK, Bruce AB, Orth DN, Geracioti TD Jr. Serial CSF corticotropin-releasing hormone levels and adrenocortical activity in combat veterans with posttraumatic stress disorder. Am J Psychiatry. 1999;156: 585–8.
29. Bremner JD, Licinio J, Darnell A, Krystal JH, Owens MJ, Southwick SM, Nemeroff CB, Charney DS. Elevated CSF corticotropin-releasing factor concentrations in posttraumatic stress disorder. Am J Psychiatry. 1997;154:624–9.
30. Chrousos GP. The hypothalamic-pituitary-adrenal axis and immune-mediated inflammation. N Engl J Med. 1995;332:1351–62.
31. Michopoulos V, Jovanovic T. Chronic inflammation: a new therapeutic target for post-traumatic stress disorder? Lancet Psychiatry. 2015;2:954–5.
32. Rosen RL, Levy-Carrick N, Reibman J, Xu N, Shao Y, Liu M, Ferri L, Kazeros A, Caplan-Shaw CE, Pradhan DR, Marmor M, Galatzer-Levy IR. Elevated C-reactive protein and posttraumatic stress pathology among survivors of the 9/11 World Trade Center attacks. J Psychiatr Res. 2017;89:14–21.
33. Solomon Z, Levin Y, Assayag EB, Furman O, Shenhar-Tsarfaty S, Berliner S, Ohry A. The implication of combat stress and PTSD trajectories in metabolic syndrome and elevated C-reactive protein levels: a longitudinal study. J Clin Psychiatry. 2017;78:e1180–e6.
34. Tursich M, Neufeld RW, Frewen PA, Harricharan S, Kibler JL, Rhind SG, Lanius RA. Association of trauma exposure with proinflammatory activity: a transdiagnostic meta-analysis. Transl Psychiatry. 2014;4:e413.

35. Passos IC, Vasconcelos-Moreno MP, Costa LG, Kunz M, Brietzke E, Quevedo J, Salum G, Magalhães PV, Kapczinski F, Kauer-Sant'Anna M. Inflammatory markers in post-traumatic stress disorder: a systematic review, meta-analysis, and meta-regression. Lancet Psychiatry. 2015;2:1002–12.
36. Trapero I, Cauli O. Interleukin 6 and cognitive dysfunction. Metab Brain Dis. 2014;29:593–608.
37. Quinones MM, Gallegos AM, Lin FV, Heffner K. Dysregulation of inflammation, neurobiology, and cognitive function in PTSD: an integrative review. Cogn Affect Behav Neurosci. 2020;20:455–80.
38. Nedic Erjavec G, Nikolac Perkovic M, Tudor L, Uzun S, Kovacic Petrovic Z, Konjevod M, Sagud M, Kozumplik O, Svob Strac D, Peraica T, Mimica N, Havelka Mestrovic A, Zilic D, Pivac N. Moderating effects of BDNF genetic variants and smoking on cognition in PTSD veterans. Biomol Ther. 2021;11:641.
39. Somvanshi PR, Mellon SH, Yehuda R, Flory JD, Makotkine I, Bierer L, Marmar C, Jett M, Doyle FJ 3rd. Role of enhanced glucocorticoid receptor sensitivity in inflammation in PTSD: insights from computational model for circadian-neuroendocrine-immune interactions. Am J Physiol Endocrinol Metab. 2020;319:E48–66.
40. Falger PR, Op den Velde W, Hovens JE, Schouten EG, De Groen JH, Van Duijn H. Current posttraumatic stress disorder and cardiovascular disease risk factors in Dutch resistance veterans from World War II. Psychother Psychosom. 1992;57:164–71.
41. Akosile W, Colquhoun D, Young R, Lawford B, Voisey J. The association between post-traumatic stress disorder and coronary artery disease: a meta-analysis. Australas Psychiatry. 2018;26:524–30.
42. Song H, Fang F, Arnberg FK, Mataix-Cols D, Fernández de la Cruz L, Almqvist C, Fall K, Lichtenstein P, Thorgeirsson G, Valdimarsdóttir UA. Stress related disorders and risk of cardiovascular disease: population based, sibling controlled cohort study. BMJ. 2019;365:l1255.
43. Vaccarino V, Goldberg J, Rooks C, Shah AJ, Veledar E, Faber TL, Votaw JR, Forsberg CW, Bremner JD. Post-traumatic stress disorder and incidence of coronary heart disease: a twin study. J Am Coll Cardiol. 2013;62:970–8.
44. Beristianos MH, Yaffe K, Cohen B, Byers AL. PTSD and risk of incident cardiovascular disease in aging veterans. Am J Geriatr Psychiatry. 2016;24:192–200.
45. Grenon SM, Owens CD, Alley H, Perez S, Whooley MA, Neylan TC, Aschbacher K, Gasper WJ, Hilton JF, Cohen BE. Posttraumatic stress disorder is associated with worse endothelial function among veterans. J Am Heart Assoc. 2016;5:e003010.
46. Howard JT, Stewart IJ, Kolaja CA, Sosnov JA, Rull RP, Torres I, Janak JC, Walker LE, Trone DW, Armenta RF. Hypertension in military veterans is associated with combat exposure and combat injury. J Hypertens. 2020;38:1293–301.
47. Jeleč V, Bajić Ž, Šimunović Filipčić I, Portolan Pajić I, Šentija Knežević M, Miloloža I, Radić-Krišto D, Benjak T, Jakšić N, Šagud M, Wang W, Filipčić I. Utilization of somatic healthcare in Croatian patients with schizophrenia spectrum disorder, major depression, PTSD and the general population. BMC Psychiatry. 2019;19:203.
48. Šagud M, Jakšić N, Vuksan-Ćusa B, Lončar M, Lončar I, Peleš AM, Miličić D, Jakovljević M. Cardiovascular disease risk factors in patients with posttraumatic stress disorder (PTSD): a narrative review. Psychiatr Danub. 2017;29:421–30.
49. Alfaddagh A, Martin SS, Leucker TM, Michos ED, Blaha MJ, Lowenstein CJ, Jones SR, Toth PP. Inflammation and cardiovascular disease: from mechanisms to therapeutics. Am J Prev Cardiol. 2020;4:100130.
50. Kapellos TS, Bonaguro L, Gemünd I, Reusch N, Saglam A, Hinkley ER, Schultze JL. Human monocyte subsets and phenotypes in major chronic inflammatory diseases. Front Immunol. 2019;10:2035.
51. Rorabaugh BR, Bui AD, Seeley SL, Eisenmann ED, Rose RM, Johnson BL, Huntley MR, Heikkila ME, Zoladz PR. Myocardial hypersensitivity to ischemic injury is not reversed by

clonidine or propranolol in a predator-based rat model of posttraumatic stress disorder. Prog Neuro-Psychopharmacol Biol Psychiatry. 2019;89:117–24.
52. Manukhina EB, Tseilikman VE, Komelkova MV, Lapshin MS, Goryacheva AV, Kondashevskaya MV, Mkhitarov VA, Lazuko SS, Tseilikman OB, Sarapultsev AP, Dmitrieva YA, Strizhikov VK, Kuzhel OP, Downey HF. Cardiac injury in rats with experimental posttraumatic stress disorder and mechanisms of its limitation in experimental posttraumatic stress disorder-resistant rats. J Appl Physiol. 1985;2021(130):759–71.
53. Nie PY, Tong L, Li MD, Fu CH, Peng JB, Ji LL. miR-142 downregulation alleviates rat PTSD-like behaviors, reduces the level of inflammatory cytokine expression and apoptosis in hippocampus, and upregulates the expression of fragile X mental retardation protein. J Neuroinflammation. 2021;18:17.
54. Pearson-Leary J, Zhao C, Bittinger K, Eacret D, Luz S, Vigderman AS, Dayanim G, Bhatnagar S. The gut microbiome regulates the increases in depressive-type behaviors and in inflammatory processes in the ventral hippocampus of stress vulnerable rats. Mol Psychiatry. 2020;25:1068–79.
55. Xue B, Xue J, Yu Y, Wei SG, Beltz TG, Felder RB, Johnson AK. Predator scent-induced sensitization of hypertension and anxiety-like behaviors. Cell Mol Neurobiol. 2022;42(4):1141–52. https://doi.org/10.1007/s10571-020-01005-y.
56. Šimić G, Tkalčić M, Vukić V, Mulc D, Španić E, Šagud M, Olucha-Bordonau FE, Vukšić M, Hof R, P. Understanding emotions: origins and roles of the amygdala. Biomol Ther. 2021;11:823.
57. Weggen JB, Darling AM, Autler AS, Hogwood AC, Decker KP, Imthurn B, Tuzzolo GM, Garten RS. Impact of acute antioxidant supplementation on vascular function and autonomic nervous system modulation in young adults with PTSD. Am J Physiol Regul Integr Comp Physiol. 2021;321:R49–61.
58. Schneider M, Schwerdtfeger A. Autonomic dysfunction in posttraumatic stress disorder indexed by heart rate variability: a meta-analysis. Psychol Med. 2020;50:1937–48.
59. Schumacher S, Niemeyer H, Engel S, Cwik JC, Laufer S, Klusmann H, Knaevelsrud C. HPA axis regulation in posttraumatic stress disorder: a meta-analysis focusing on potential moderators. Neurosci Biobehav Rev. 2019;100:35–57.
60. Nijm J, Jonasson L. Inflammation and cortisol response in coronary artery disease. Ann Med. 2009;41:224–33.
61. Sumner JA, Maihofer AX, Michopoulos V, Rothbaum AO, Almli LM, Andreassen OA, Ashley-Koch AE, Baker DG, Beckham JC, Bradley B, Breen G, Coleman JRI, Dale AM, Dennis MF, Feeny NC, Franz CE, Garrett ME, Gillespie CF, Guffanti G, Hauser MA, Hemmings SMJ, Jovanovic T, Kimbrel NA, Kremen WS, Lawford BR, Logue MW, Lori A, Lyons MJ, Maples-Keller J, Mavissakalian MR, McGlinchey RE, Mehta D, Mellor R, Milberg W, Miller MW, Morris CP, Panizzon MS, Ressler KJ, Risbrough VB, Rothbaum BO, Roy-Byrne P, Seedat S, Smith AK, Stevens JS, van den Heuvel LL, Voisey J, Young RM, Zoellner LA, Nievergelt CM, Wolf EJ. Examining individual and synergistic contributions of PTSD and genetics to blood pressure: a trans-ethnic meta-analysis. Front Neurosci. 2021;15:678503.
62. Bierhaus A, Wolf J, Andrassy M, Rohleder N, Humpert PM, Petrov D, Ferstl R, von Eynatten M, Wendt T, Rudofsky G, Joswig M, Morcos M, Schwaninger M, McEwen B, Kirschbaum C, Nawroth PP. A mechanism converting psychosocial stress into mononuclear cell activation. Proc Natl Acad Sci U S A. 2003;100:1920–5.
63. Bremner JD, Gurel NZ, Wittbrodt MT, Shandhi MH, Rapaport MH, Nye JA, Pearce BD, Vaccarino V, Shah AJ, Park J, Bikson M, Inan OT. Application of noninvasive vagal nerve stimulation to stress-related psychiatric disorders. J Pers Med. 2020;10:119.
64. Fonkoue IT, Michopoulos V, Park J. Sex differences in post-traumatic stress disorder risk: autonomic control and inflammation. Clin Auton Res. 2020;30:409–21.
65. Pope BS, Wood SK. Stress-induced inflammation as the "connexin" between post-traumatic stress disorder and cardiovascular disease. Brain Behav Immun. 2019;82:3–5.

66. Fonkoue IT, Marvar PJ, Norrholm S, Li Y, Kankam ML, Jones TN, Vemulapalli M, Rothbaum B, Bremner JD, Le NA, Park J. Symptom severity impacts sympathetic dysregulation and inflammation in post-traumatic stress disorder (PTSD). Brain Behav Immun. 2020;83:260–9.
67. Lima BB, Hammadah M, Wilmot K, Pearce BD, Shah A, Levantsevych O, Kaseer B, Obideen M, Gafeer MM, Kim JH, Sullivan S, Lewis TT, Weng L, Elon L, Li L, Bremner JD, Raggi P, Quyyumi A, Vaccarino V. Posttraumatic stress disorder is associated with enhanced interleukin-6 response to mental stress in subjects with a recent myocardial infarction. Brain Behav Immun. 2019;75:26–33. Erratum in: Brain Behav Immun. 2019;78:204–5.
68. Koraishy FM, Salas J, Neylan TC, Cohen BE, Schnurr PP, Clouston S, Scherrer JF. Association of severity of posttraumatic stress disorder with inflammation: using total white blood cell count as a marker. Chron Stress (Thousand Oaks). 2019;3: 2470547019877651.
69. Kim TD, Lee S, Yoon S. Inflammation in post-traumatic stress disorder (PTSD): a review of potential correlates of PTSD with a neurological perspective. Antioxidants (Basel). 2020;9: 107.
70. Escarfulleri S, Ellickson-Larew S, Fein-Schaffer D, Mitchell KS, Wolf EJ. Emotion regulation and the association between PTSD, diet, and exercise: a longitudinal evaluation among US military veterans. Eur J Psychotraumatol. 2021;12:1895515.
71. Battineni G, Sagaro GG, Chintalapudi N, Amenta F, Tomassoni D, Tayebati SK. Impact of obesity-induced inflammation on cardiovascular diseases (CVD). Int J Mol Sci. 2021;22:4798.
72. van den Berk-Clark C, Secrest S, Walls J, Hallberg E, Lustman PJ, Schneider FD, Scherrer JF. Association between posttraumatic stress disorder and lack of exercise, poor diet, obesity, and co-occuring smoking: a systematic review and meta-analysis. Health Psychol. 2018;37: 407–16.
73. Tudor L, Konjevod M, Nikolac Perkovic M, Svob Strac D, Nedic Erjavec G, Uzun S, Kozumplik O, Sagud M, Kovacic Petrovic Z, Pivac N. Genetic variants of the brain-derived neurotrophic factor and metabolic indices in veterans with posttraumatic stress disorder. Front Psych. 2018;9:637.
74. Šagud M, Petrović B, Vilibić M, Mihaljević-Peleš A, Vuksan-Ćusa B, Radoš I, Greš A, Trkulja V. The relationship among psychopathology, religiosity, and nicotine dependence in Croatian war veterans with posttraumatic stress disorder. Croat Med J. 2018;59:165–77.
75. Dennis PA, Weinberg JB, Calhoun PS, Watkins LL, Sherwood A, Dennis MF, Beckham JC. An investigation of Vago-regulatory and health-behavior accounts for increased inflammation in posttraumatic stress disorder. J Psychosom Res. 2016;83:33–9.
76. Goetz M, Shah A, Goldberg J, Cheema F, Shallenberger L, Murrah NV, Bremner JD, Vaccarino V. Posttraumatic stress disorder, combat exposure, and carotid intima-media thickness in male twins. Am J Epidemiol. 2014;180:989–96.
77. Rhein C, Hepp T, Kraus O, von Majewski K, Lieb M, Rohleder N, Erim Y. Interleukin-6 secretion upon acute psychosocial stress as a potential predictor of psychotherapy outcome in posttraumatic stress disorder. J Neural Transm (Vienna). 2021;128:1301–10.
78. McD Young R, Lawford B, Mellor R, Morris CP, Voisey J, Initiative PTSD. Investigation of C-reactive protein and AIM2 methylation as a marker for PTSD in Australian Vietnam veterans. Gene. 2021;803:145898.
79. Zhang JM, An J. Cytokines, inflammation, and pain. Int Anesthesiol Clin. 2007;45:27–37.
80. Vezzani A, Viviani B. Neuromodulatory properties of inflammatory cytokines and their impact on neuronal excitability. Neuropharmacology. 2015;96:70–82.
81. Levin SG, Godukhin OV. Modulating effect of cytokines on mechanisms of synaptic plasticity in the brain. Biochemistry (Mosc). 2017;82:264–74.
82. Vezzani A, Maroso M, Balosso S, Sanchez MA, Bartfai T. IL-1 receptor/toll-like receptor signaling in infection, inflammation, stress and neurodegeneration couples hyperexcitability and seizures. Brain Behav Immun. 2011;25:1281–9.

83. Tan KS, Nackley AG, Satterfield K, Maixner W, Diatchenko L, Flood PM. Beta2 adrenergic receptor activation stimulates pro-inflammatory cytokine production in macrophages via PKA- and NF-kappaB-independent mechanisms. Cell Signal. 2007;19:251–60.
84. Bruenig D, Mehta D, Morris CP, Harvey W, Lawford B, Young RM, Voisey J. Genetic and serum biomarker evidence for a relationship between TNFα and PTSD in Vietnam war combat veterans. Compr Psychiatry. 2017;74:125–33.
85. Yang JJ, Jiang W. Immune biomarkers alterations in post-traumatic stress disorder: a systematic review and meta-analysis. J Affect Disord. 2020;268:39–46.
86. Danan ER, Krebs EE, Ensrud K, Koeller E, MacDonald R, Velasquez T, Greer N, Wilt TJ. An evidence map of the women veterans' health research literature (2008-2015). J Gen Intern Med. 2017;32:1359–76.
87. Lalonde CS, Mekawi Y, Ethun KF, Beurel E, Gould F, Dhabhar FS, Schultebraucks K, Galatzer-Levy I, Maples-Keller JL, Rothbaum BO, Ressler KJ, Nemeroff CB, Stevens JS, Michopoulos V. Sex differences in peritraumatic inflammatory cytokines and steroid hormones contribute to prospective risk for nonremitting posttraumatic stress disorder. Chron Stress (Thousand Oaks). 2021;5:24705470211032208.
88. Miller AH, Raison CL. The role of inflammation in depression: from evolutionary imperative to modern treatment target. Nat Rev Immunol. 2016;16:22–34.
89. Baumeister D, Akhtar R, Ciufolini S, Pariante CM, Mondelli V. Childhood trauma and adulthood inflammation: a meta-analysis of peripheral C-reactive protein, interleukin-6 and tumour necrosis factor-α. Mol Psychiatry. 2016;21:642–9.
90. Van Praag DLG, Cnossen MC, Polinder S, Wilson L, Maas AIR. Post-traumatic stress disorder after civilian traumatic brain injury: a systematic review and meta-analysis of prevalence rates. J Neurotrauma. 2019;36:3220–32.
91. Rodney T, Taylor P, Dunbar K, Perrin N, Lai C, Roy M, Gill J. High IL-6 in military personnel relates to multiple traumatic brain injuries and post-traumatic stress disorder. Behav Brain Res. 2020;392:112715.
92. Katerji M, Filippova M, Duerksen-Hughes P. Approaches and methods to measure oxidative stress in clinical samples: research applications in the cancer field. Oxidative Med Cell Longev. 2019;2019:1279250.
93. Schiavone S, Colaianna M, Curtis L. Impact of early life stress on the pathogenesis of mental disorders: relation to brain oxidative stress. Curr Pharm Des. 2015;21:1404–12.
94. Borovac Štefanović L, Kalinić D, Mimica N, Beer Ljubić B, Aladrović J, Mandelsamen Perica M, Curić M, Grošić PF, Delaš I. Oxidative status and the severity of clinical symptoms in patients with post-traumatic stress disorder. Ann Clin Biochem. 2015;52:95–104.
95. Tezcan E, Atmaca M, Kuloglu M, Ustundag B. Free radicals in patients with post-traumatic stress disorder. Eur Arch Psychiatry Clin Neurosci. 2003;253:89–91.
96. Attari A, Asgari S, Naderi GA, Rezayat A. Lipid peroxidation and antioxidant capacity in posttraumatic stress disorder. J Isfahan Med Sch. 2002;20:4–6.
97. Atli A, Bulut M, Bez Y, Kaplan İ, Özdemir PG, Uysal C, Selçuk H, Sir A. Altered lipid peroxidation markers are related to post-traumatic stress disorder (PTSD) and not trauma itself in earthquake survivors. Eur Arch Psychiatry Clin Neurosci. 2016;266:329–36.
98. Perković MN, Milković L, Uzun S, Mimica N, Pivac N, Waeg G, Žarković N. Association of lipid peroxidation product 4-hydroxynonenal with post-traumatic stress disorder. Biomol Ther. 2021;11:1365.
99. Michels L, Schulte-Vels T, Schick M, O'Gorman RL, Zeffiro T, Hasler G, Mueller-Pfeiffer C. Prefrontal GABA and glutathione imbalance in posttraumatic stress disorder: preliminary findings. Psychiatry Res. 2014;224:288–95.
100. Glatt SJ, Tylee DS, Chandler SD, Pazol J, Nievergelt CM, Woelk CH, Baker DG, Lohr JB, Kremen WS, Litz BT, Tsuang MT, Marine Resiliency Study Investigators. Blood-based gene-expression predictors of PTSD risk and resilience among deployed marines: a pilot study. Am J Med Genet B Neuropsychiatr Genet. 2013;162B:313–26.

101. Tylee DS, Chandler SD, Nievergelt CM, Liu X, Pazol J, Woelk CH, Lohr JB, Kremen WS, Baker DG, Glatt SJ, Tsuang MT. Marine resiliency study investigators. Blood-based gene-expression biomarkers of post-traumatic stress disorder among deployed marines: a pilot study. Psychoneuroendocrinology. 2015;51:472–94.
102. Konjevod M, Nedic Erjavec G, Nikolac Perkovic M, Sáiz J, Tudor L, Uzun S, Kozumplik O, Svob Strac D, Zarkovic N, Pivac N. Metabolomics in posttraumatic stress disorder: untargeted metabolomic analysis of plasma samples from Croatian war veterans. Free Radic Biol Med. 2021;162:636–41.
103. Ceprnja M, Derek L, Unić A, Blazev M, Fistonić M, Kozarić-Kovacić D, Franić M, Romić Z. Oxidative stress markers in patients with post-traumatic stress disorder. Coll Antropol. 2011;35:1155–60.
104. Şimşek Ş, Yüksel T, Kaplan İ, Uysal C, Aktaş H. The levels of cortisol and oxidative stress and DNA damage in child and adolescent victims of sexual abuse with or without post-traumatic stress disorder. Psychiatry Investig. 2016;13:616–21.
105. Gulec M, Ozkol H, Selvi Y, Tuluce Y, Aydin A, Besiroglu L, Ozdemir PG. Oxidative stress in patients with primary insomnia. Prog Neuro-Psychopharmacol Biol Psychiatry. 2012;37:247–51.
106. Silva RH, Abílio VC, Takatsu AL, Kameda SR, Grassl C, Chehin AB, Medrano WA, Calzavara MB, Registro S, Andersen ML, Machado RB, Carvalho RC, Ribeiro Rde A, Tufik S, Frussa-Filho R. Role of hippocampal oxidative stress in memory deficits induced by sleep deprivation in mice. Neuropharmacology. 2004;46:895–903.
107. Mullington JM, Simpson NS, Meier-Ewert HK, Haack M. Sleep loss and inflammation. Best Pract Res Clin Endocrinol Metab. 2010;24:775–84.
108. Hurtado-Alvarado G, Pavón L, Castillo-García SA, Hernández ME, Domínguez-Salazar E, Velázquez-Moctezuma J, Gómez-González B. Sleep loss as a factor to induce cellular and molecular inflammatory variations. Clin Dev Immunol. 2013;2013:801341.
109. Laing KJ, Secombes CJ. Chemokines. Dev Comp Immunol. 2004;28:443–60.
110. Shams K, Kurowska-Stolarska M, Schütte F, Burden AD, McKimmie CS, Graham GJ. MicroRNA-146 and cell trauma down-regulate expression of the psoriasis-associated atypical chemokine receptor ACKR2. J Biol Chem. 2018;293:3003–12.
111. Ogłodek EA, Szota AM, Moś DM, Araszkiewicz A, Szromek AR. Serum concentrations of chemokines (CCL-5 and CXCL-12), chemokine receptors (CCR-5 and CXCR-4), and IL-6 in patients with posttraumatic stress disorder and avoidant personality disorder. Pharmacol Rep. 2015;67:1251–8.
112. Ogłodek EA, Szota AM, Just MJ, Szromek AR, Araszkiewicz A. A study of chemokines, chemokine receptors and interleukin-6 in patients with panic disorder, personality disorders and their co-morbidity. Pharmacol Rep. 2016;68:756–63.
113. Kim YK, Amidfar M, Won E. A review on inflammatory cytokine-induced alterations of the brain as potential neural biomarkers in post-traumatic stress disorder. Prog Neuro-Psychopharmacol Biol Psychiatry. 2019;91:103–12.
114. Pan X, Kaminga AC, Wu Wen S, Liu A. Chemokines in post-traumatic stress disorder: a network meta-analysis. Brain Behav Immun. 2021;92:115–26.
115. Zhang L, Hu XZ, Li X, Chen Z, Benedek DM, Fullerton CS, Wynn G, Biomarker team, Ursano RJ. Potential chemokine biomarkers associated with PTSD onset, risk and resilience as well as stress responses in US military service members. Transl. Psychiatry. 2020;10:31.
116. Toft H, Bramness JG, Lien L, Abebe DS, Wampold BE, Tilden T, Hestad K, Neupane SP. PTSD patients show increasing cytokine levels during treatment despite reduced psychological distress. Neuropsychiatr Dis Treat. 2018;14:2367–78.
117. Friend SF, Nachnani R, Powell SB, Risbrough VB. C-reactive protein: marker of risk for post-traumatic stress disorder and its potential for a mechanistic role in trauma response and recovery. Eur J Neurosci. 2020;55(9–10):2297–310. https://doi.org/10.1111/ejn.15031.
118. Pepys MB, Hirschfield GM. C-reactive protein: a critical update. J Clin Invest. 2003;111:1805–12.

119. Volanakis JE. Human C-reactive protein: expression, structure, and function. Mol Immunol. 2001;38:189–97.
120. Singh U, Devaraj S, Dasu MR, Ciobanu D, Reusch J, Jialal I. C-reactive protein decreases interleukin-10 secretion in activated human monocyte-derived macrophages via inhibition of cyclic AMP production. Arterioscler Thromb Vasc Biol. 2006;26:2469–75.
121. Koziarska-Rościszewska M, Gluba-Brzózka A, Franczyk B, Rysz J. High-sensitivity C-reactive protein relationship with metabolic disorders and cardiovascular diseases risk factors. Life (Basel). 2021;11:742.
122. Söndergaard HP, Hansson LO, Theorell T. The inflammatory markers C-reactive protein and serum amyloid a in refugees with and without posttraumatic stress disorder. Clin Chim Acta. 2004;342:93–8.
123. Breen MS, Tylee DS, Maihofer AX, Neylan TC, Mehta D, Binder EB, Chandler SD, Hess JL, Kremen WS, Risbrough VB, Woelk CH, Baker DG, Nievergelt CM, Tsuang MT, Buxbaum JD, Glatt SJ. PTSD blood transcriptome mega-analysis: shared inflammatory pathways across biological sex and modes of trauma. Neuropsychopharmacology. 2018;43:469–81.
124. Groer MW, Kane B, Williams SN, Duffy A. Relationship of PTSD symptoms with combat exposure, stress, and inflammation in American soldiers. Biol Res Nurs. 2015;17:303–10.
125. Spitzer C, Barnow S, Völzke H, Wallaschofski H, John U, Freyberger HJ, Löwe B, Grabe HJ. Association of posttraumatic stress disorder with low-grade elevation of C-reactive protein: evidence from the general population. J Psychiatr Res. 2010;44:15–21.
126. Sumner JA, Chen Q, Roberts AL, Winning A, Rimm EB, Gilsanz P, Glymour MM, Tworoger SS, Koenen KC, Kubzansky LD. Cross-sectional and longitudinal associations of chronic posttraumatic stress disorder with inflammatory and endothelial function markers in women. Biol Psychiatry. 2017;82:875–84.
127. Di Napoli M, Godoy DA, Campi V, Masotti L, Smith CJ, Parry Jones AR, Hopkins SJ, Slevin M, Papa F, Mogoanta L, Pirici D, Popa Wagner A. C-reactive protein in intracerebral hemorrhage: time course, tissue localization, and prognosis. Neurology. 2012;79:690–9.
128. Iwamoto N, Nishiyama E, Ohwada J, Arai H. Demonstration of CRP immunoreactivity in brains of Alzheimer's disease: immunohistochemical study using formic acid pretreatment of tissue sections. Neurosci Lett. 1994;177:23–6.
129. Strang F, Scheichl A, Chen YC, Wang X, Htun NM, Bassler N, Eisenhardt SU, Habersberger J, Peter K. Amyloid plaques dissociate pentameric to monomeric C-reactive protein: a novel pathomechanism driving cortical inflammation in Alzheimer's disease? Brain Pathol. 2012;22:337–46.
130. Yasojima K, Schwab C, McGeer EG, McGeer PL. Human neurons generate C-reactive protein and amyloid P: upregulation in Alzheimer's disease. Brain Res. 2000;887:80–9.
131. Felger JC, Li Z, Haroon E, Woolwine BJ, Jung MY, Hu X, Miller AH. Inflammation is associated with decreased functional connectivity within corticostriatal reward circuitry in depression. Mol Psychiatry. 2016;21:1358–65.
132. Haroon E, Chen X, Li Z, Patel T, Woolwine BJ, Hu XP, Felger JC, Miller AH. Increased inflammation and brain glutamate define a subtype of depression with decreased regional homogeneity, impaired network integrity, and anhedonia. Transl Psychiatry. 2018;8:189.
133. Alexandrov PN, Kruck TP, Lukiw WJ. Nanomolar aluminum induces expression of the inflammatory systemic biomarker C-reactive protein (CRP) in human brain microvessel endothelial cells (hBMECs). J Inorg Biochem. 2015;152:210–3.
134. Elwood E, Lim Z, Naveed H, Galea I. The effect of systemic inflammation on human brain barrier function. Brain Behav Immun. 2017;62:35–40.
135. Prakash R, Carmichael ST. Blood-brain barrier breakdown and neovascularization processes after stroke and traumatic brain injury. Curr Opin Neurol. 2015;28:556–64.
136. Welcome MO, Mastorakis NE. Stress-induced blood brain barrier disruption: molecular mechanisms and signaling pathways. Pharmacol Res. 2020;157:104769.

137. Eraly SA, Nievergelt CM, Maihofer AX, Barkauskas DA, Biswas N, Agorastos A, O'Connor DT, Baker DG, Marine Resiliency Study Team. Assessment of plasma C-reactive protein as a biomarker of posttraumatic stress disorder risk. JAMA Psychiat. 2014;71:423–31.
138. Sumner JA, Chen Q, Roberts AL, Winning A, Rimm EB, Gilsanz P, Glymour MM, Tworoger SS, Koenen KC, Kubzansky LD. Posttraumatic stress disorder onset and inflammatory and endothelial function biomarkers in women. Brain Behav Immun. 2018;69:203–9.
139. Khera A, McGuire DK, Murphy SA, Stanek HG, Das SR, Vongpatanasin W, Wians FH Jr, Grundy SM, de Lemos JA. Race and gender differences in C-reactive protein levels. J Am Coll Cardiol. 2005;46:464–9.
140. Lee S, Oh SS, Jang SI, Park EC. Sex difference in the association between high-sensitivity C-reactive protein and depression: the 2016 Korea National Health and Nutrition Examination Survey. Sci Rep. 2019;9:1918.
141. Heath NM, Chesney SA, Gerhart JI, Goldsmith RE, Luborsky JL, Stevens NR, Hobfoll SE. Interpersonal violence, PTSD, and inflammation: potential psychogenic pathways to higher C-reactive protein levels. Cytokine. 2013;63:172–8.
142. Miller RJ, Sutherland AG, Hutchison JD, Alexander DA. C-reactive protein and interleukin 6 receptor in post-traumatic stress disorder: a pilot study. Cytokine. 2001;13:253–5.
143. Gillespie CF, Bradley B, Mercer K, Smith AK, Conneely K, Gapen M, Weiss T, Schwartz AC, Cubells JF, Ressler KJ. Trauma exposure and stress-related disorders in inner city primary care patients. Gen Hosp Psychiatry. 2009;31:505–14.
144. Melamed S, Shirom A, Toker S, Berliner S, Shapira I. Association of fear of terror with low-grade inflammation among apparently healthy employed adults. Psychosom Med. 2004;66:484–91.
145. Relja B, Land WG. Damage-associated molecular patterns in trauma. Eur J Trauma Emerg Surg. 2020;46:751–75.
146. Bertone-Johnson ER, Whitcomb BW, Missmer SA, Karlson EW, Rich-Edwards JW. Inflammation and early-life abuse in women. Am J Prev Med. 2012;43:611–20.
147. Lin JE, Neylan TC, Epel E, O'Donovan A. Associations of childhood adversity and adulthood trauma with C-reactive protein: a cross-sectional population-based study. Brain Behav Immun. 2016;53:105–12.
148. O'Donovan A, Neylan TC, Metzler T, Cohen BE. Lifetime exposure to traumatic psychological stress is associated with elevated inflammation in the Heart and Soul Study. Brain Behav Immun. 2012;26:642–9.
149. Edwards VJ, Holden GW, Felitti VJ, Anda RF. Relationship between multiple forms of childhood maltreatment and adult mental health in community respondents: results from the adverse childhood experiences study. Am J Psychiatry. 2003;160:1453–60.
150. Parto JA, Evans MK, Zonderman AB. Symptoms of posttraumatic stress disorder among urban residents. J Nerv Ment Dis. 2011;199:436–9.
151. Nazmi A, Victora CG. Socioeconomic and racial/ethnic differentials of C-reactive protein levels: a systematic review of population-based studies. BMC Public Health. 2007;7:212.
152. Liu RS, Aiello AE, Mensah FK, Gasser CE, Rueb K, Cordell B, Juonala M, Wake M, Burgner DP. Socioeconomic status in childhood and C reactive protein in adulthood: a systematic review and meta-analysis. J Epidemiol Community Health. 2017;71:817–26.
153. Stringhini S, Polidoro S, Sacerdote C, Kelly RS, van Veldhoven K, Agnoli C, Grioni S, Tumino R, Giurdanella MC, Panico S, Mattiello A, Palli D, Masala G, Gallo V, Castagné R, Paccaud F, Campanella G, Chadeau-Hyam M, Vineis P. Life-course socioeconomic status and DNA methylation of genes regulating inflammation. Int J Epidemiol. 2015;44:1320–30.
154. De Nutte L, Okello J, Derluyn I. Social relationships and social support among post-war youth in Northern Uganda. Int J Psychol. 2017;52:291–9.
155. Lee JS. Perceived social support functions as a resilience in buffering the impact of trauma exposure on PTSD symptoms via intrusive rumination and entrapment in firefighters. PLoS One. 2019;14:e0220454.

156. Elliot AJ, Heffner KL, Mooney CJ, Moynihan JA, Chapman BP. Social relationships and inflammatory markers in the MIDUS cohort: the role of age and gender differences. J Aging Health. 2018;30:904–23.
157. Shimanoe C, Hara M, Nishida Y, Nanri H, Otsuka Y, Horita M, Yasukata J, Miyoshi N, Yamada Y, Higaki Y, Tanaka K. Coping strategy and social support modify the association between perceived stress and C-reactive protein: a longitudinal study of healthy men and women. Stress. 2018;21:237–46.
158. Ligthart S, Vaez A, Võsa U, Stathopoulou MG, de Vries PS, Prins BP, Van der Most PJ, Tanaka T, Naderi E, Rose LM, Wu Y, Karlsson R, Barbalic M, Lin H, Pool R, Zhu G, Macé A, Sidore C, Trompet S, Mangino M, Sabater-Lleal M, Kemp JP, Abbasi A, Kacprowski T, Verweij N, Smith AV, Huang T, Marzi C, Feitosa MF, Lohman KK, Kleber ME, Milaneschi Y, Mueller C, Huq M, Vlachopoulou E, Lyytikäinen LP, Oldmeadow C, Deelen J, Perola M, Zhao JH, Feenstra B, LifeLines Cohort Study, Amini M, CHARGE Inflammation Working Group, Lahti J, Schraut KE, Fornage M, Suktitipat B, Chen WM, Li X, Nutile T, Malerba G, Luan J, Bak T, Schork N, Del Greco MF, Thiering E, Mahajan A, Marioni RE, Mihailov E, Eriksson J, Ozel AB, Zhang W, Nethander M, Cheng YC, Aslibekyan S, Ang W, Gandin I, Yengo L, Portas L, Kooperberg C, Hofer E, Rajan KB, Schurmann C, den Hollander W, Ahluwalia TS, Zhao J, Draisma HHM, Ford I, Timpson N, Teumer A, Huang H, Wahl S, Liu Y, Huang J, Uh HW, Geller F, Joshi PK, Yanek LR, Trabetti E, Lehne B, Vozzi D, Verbanck M, Biino G, Saba Y, Meulenbelt I, O'Connell JR, Laakso M, Giulianini F, Magnusson PKE, Ballantyne CM, Hottenga JJ, Montgomery GW, Rivadineira F, Rueedi R, Steri M, Herzig KH, Stott DJ, Menni C, Frånberg M, St Pourcain B, Felix SB, Pers TH, Bakker SJL, Kraft P, Peters A, Vaidya D, Delgado G, Smit JH, Großmann V, Sinisalo J, Seppälä I, Williams SR, Holliday EG, Moed M, Langenberg C, Räikkönen K, Ding J, Campbell H, Sale MM, Chen YI, James AL, Ruggiero D, Soranzo N, Hartman CA, Smith EN, Berenson GS, Fuchsberger C, Hernandez D, Tiesler CMT, Giedraitis V, Liewald D, Fischer K, Mellström D, Larsson A, Wang Y, Scott WR, Lorentzon M, Beilby J, Ryan KA, Pennell CE, Vuckovic D, Balkau B, Concas MP, Schmidt R, Mendes de Leon CF, Bottinger EP, Kloppenburg M, Paternoster L, Boehnke M, Musk AW, Willemsen G, Evans DM, Madden PAF, Kähönen M, Kutalik Z, Zoledziewska M, Karhunen V, Kritchevsky SB, Sattar N, Lachance G, Clarke R, Harris TB, Raitakari OT, Attia JR, van Heemst D, Kajantie E, Sorice R, Gambaro G, Scott RA, Hicks AA, Ferrucci L, Standl M, Lindgren CM, Starr JM, Karlsson M, Lind L, Li JZ, Chambers JC, Mori TA, de Geus EJCN, Heath AC, Martin NG, Auvinen J, Buckley BM, de Craen AJM, Waldenberger M, Strauch K, Meitinger T, Scott RJ, McEvoy M, Beekman M, Bombieri C, Ridker PM, Mohlke KL, Pedersen NL, Morrison AC, Boomsma DI, Whitfield JB, Strachan DP, Hofman A, Vollenweider P, Cucca F, Jarvelin MR, Jukema JW, Spector TD, Hamsten A, Zeller T, Uitterlinden AG, Nauck M, Gudnason V, Qi L, Grallert H, Borecki IB, Rotter JI, März W, Wild PS, Lokki ML, Boyle M, Salomaa V, Melbye M, Eriksson JG, Wilson JF, Penninx BWJH, Becker DM, Worrall BB, Gibson G, Krauss RM, Ciullo M, Zaza G, Wareham NJ, Oldehinkel AJ, Palmer LJ, Murray SS, Pramstaller PP, Bandinelli S, Heinrich J, Ingelsson E, Deary IJ, Mägi R, Vandenput L, van der Harst P, Desch KC, Kooner JS, Ohlsson C, Hayward C, Lehtimäki T, Shuldiner AR, Arnett DK, Beilin LJ, Robino A, Froguel P, Pirastu M, Jess T, Koenig W, Loos RJF, Evans DA, Schmidt H, Smith GD, Slagboom PE, Eiriksdottir G, Morris AP, Psaty BM, Tracy RP, Nolte IM, Boerwinkle E, Visvikis-Siest S, Reiner AP, Gross M, Bis JC, Franke L, Franco OH, Benjamin EJ, Chasman DI, Dupuis J, Snieder H, Dehghan A, Alizadeh BZ. Genome analyses of >200,000 individuals identify 58 loci for chronic inflammation and highlight pathways that link inflammation and complex disorders. Am J Hum Genet. 2018;103:691–706.
159. Almeida OP, Norman PE, Allcock R, van Bockxmeer F, Hankey GJ, Jamrozik K, Flicker L. Polymorphisms of the CRP gene inhibit inflammatory response and increase susceptibility to depression: the Health in Men Study. Int J Epidemiol. 2009;38:1049–59.

160. Szalai AJ, Wu J, Lange EM, McCrory MA, Langefeld CD, Williams A, Zakharkin SO, George V, Allison DB, Cooper GS, Xie F, Fan Z, Edberg JC, Kimberly RP. Single-nucleotide polymorphisms in the C-reactive protein (CRP) gene promoter that affect transcription factor binding, alter transcriptional activity, and associate with differences in baseline serum CRP level. J Mol Med (Berl). 2005;83:440–7.
161. Hernández-Díaz Y, Tovilla-Zárate CA, Juárez-Rojop I, Baños-González MA, Torres-Hernández ME, López-Narváez ML, Yañez-Rivera TG, González-Castro TB. The role of gene variants of the inflammatory markers CRP and TNF-α in cardiovascular heart disease: systematic review and meta-analysis. Int J Clin Exp Med. 2015;8:11958–84.
162. Miller MW, Maniates H, Wolf EJ, Logue MW, Schichman SA, Stone A, Milberg W, McGlinchey R. CRP polymorphisms and DNA methylation of the AIM2 gene influence associations between trauma exposure, PTSD, and C-reactive protein. Brain Behav Immun. 2018;67:194–202.
163. Fernandes-Alnemri T, Yu JW, Datta P, Wu J, Alnemri ES. AIM2 activates the inflammasome and cell death in response to cytoplasmic DNA. Nature. 2009;458:509–13.
164. Muniz Carvalho C, Wendt FR, Maihofer AX, Stein DJ, Stein MB, Sumner JA, Hemmings SMJ, Nievergelt CM, Koenen KC, Gelernter J, Belangero SI, Polimanti R. Dissecting the genetic association of C-reactive protein with PTSD, traumatic events, and social support. Neuropsychopharmacology. 2021;46:1071–7.
165. Chen Y, Guillemin GJ. Kynurenine pathway metabolites in humans: disease and healthy states. Int J Tryptophan Res. 2009;2:1–19.
166. Schwarcz R, Bruno JP, Muchowski PJ, Wu HQ. Kynurenines in the mammalian brain: when physiology meets pathology. Nat Rev Neurosci. 2012;13:465–77.
167. Cervenka I, Agudelo LZ, Ruas JL. Kynurenines: Tryptophan's metabolites in exercise, inflammation, and mental health. Science. 2017;357(6349):eaaf9794.
168. O'Farrell K, Fagan E, Connor TJ, Harkin A. Inhibition of the kynurenine pathway protects against reactive microglial-associated reductions in the complexity of primary cortical neurons. Eur J Pharmacol. 2017;810:163–73.
169. Mithaiwala MN, Santana-Coelho D, Porter GA, O'Connor JC. Neuroinflammation and the kynurenine pathway in CNS disease: molecular mechanisms and therapeutic implications. Cell. 2021;10:1548.
170. Myint AM. Kynurenines: from the perspective of major psychiatric disorders. FEBS J. 2012;279:1375–85.
171. O'Farrell K, Harkin A. Stress-related regulation of the kynurenine pathway: relevance to neuropsychiatric and degenerative disorders. Neuropharmacology. 2017;112:307–23.
172. Lovelace MD, Varney B, Sundaram G, Lennon MJ, Lim CK, Jacobs K, Guillemin GJ, Brew BJ. Recent evidence for an expanded role of the kynurenine pathway of tryptophan metabolism in neurological diseases. Neuropharmacology. 2017;112:373–88.
173. Schwarcz R, Stone TW. The kynurenine pathway and the brain: challenges, controversies and promises. Neuropharmacology. 2017;112:237–47.
174. Kim YK, Jeon SW. Neuroinflammation and the immune-kynurenine pathway in anxiety disorders. Curr Neuropharmacol. 2018;16:574–82.
175. Tanaka M, Tóth F, Polyák H, Szabó Á, Mándi Y, Vécsei L. Immune influencers in action: metabolites and enzymes of the tryptophan-kynurenine metabolic pathway. Biomedicine. 2021;9:734.
176. Badawy AA. Kynurenine pathway of tryptophan metabolism: regulatory and functional aspects. Int J Tryptophan Res. 2017;10:1178646917691938.
177. Maes M, Leonard BE, Myint AM, Kubera M, Verkerk R. The new '5-HT' hypothesis of depression: cell-mediated immune activation induces indoleamine 2,3-dioxygenase, which leads to lower plasma tryptophan and an increased synthesis of detrimental tryptophan catabolites (TRYCATs), both of which contribute to the onset of depression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2011;35:702–21.

178. Guillemin GJ, Cullen KM, Lim CK, Smythe GA, Garner B, Kapoor V, Takikawa O, Brew BJ. Characterization of the kynurenine pathway in human neurons. J Neurosci. 2007;27: 12884–92.
179. Moffett JR, Namboodiri MA. Tryptophan and the immune response. Immunol Cell Biol. 2003;81:247–65.
180. Barone P. The 'Yin' and the 'Yang' of the kynurenine pathway: excitotoxicity and neuroprotection imbalance in stress-induced disorders. Behav Pharmacol. 2019;30:163–86.
181. Guillemin GJ. Quinolinic acid, the inescapable neurotoxin. FEBS J. 2012;279:1356–65.
182. Lugo-Huitrón R, Blanco-Ayala T, Ugalde-Muñiz P, Carrillo-Mora P, Pedraza-Chaverrí J, Silva-Adaya D, Maldonado PD, Torres I, Pinzón E, Ortiz-Islas E, López T, García E, Pineda B, Torres-Ramos M, Santamaría A, La Cruz VP. On the antioxidant properties of kynurenic acid: free radical scavenging activity and inhibition of oxidative stress. Neurotoxicol Teratol. 2011;33:538–47.
183. Wirthgen E, Hoeflich A, Rebl A, Günther J. Kynurenic acid: the Janus-faced role of an immunomodulatory tryptophan metabolite and its link to pathological conditions. Front Immunol. 2018;8:1957.
184. Jhamandas KH, Boegman RJ, Beninger RJ, Miranda AF, Lipic KA. Excitotoxicity of quinolinic acid: modulation by endogenous antagonists. Neurotox Res. 2000;2:139–55.
185. Konradsson-Geuken A, Wu HQ, Gash CR, Alexander KS, Campbell A, Sozeri Y, Pellicciari R, Schwarcz R, Bruno JP. Cortical kynurenic acid bi-directionally modulates prefrontal glutamate levels as assessed by microdialysis and rapid electrochemistry. Neuroscience. 2010;169:1848–59.
186. Moroni F, Cozzi A, Sili M, Mannaioni G. Kynurenic acid: a metabolite with multiple actions and multiple targets in brain and periphery. J Neural Transm (Vienna). 2012;119:133–9.
187. Ferreira FS, Biasibetti-Brendler H, Pierozan P, Schmitz F, Bertó CG, Prezzi CA, Manfredini V, Wyse ATS. Kynurenic acid restores Nrf2 levels and prevents quinolinic acid-induced toxicity in rat striatal slices. Mol Neurobiol. 2018;55:8538–49.
188. Dounay AB, Tuttle JB, Verhoest PR. Challenges and opportunities in the discovery of new therapeutics targeting the kynurenine pathway. J Med Chem. 2015;58:8762–82.
189. Campbell BM, Charych E, Lee AW, Möller T. Kynurenines in CNS disease: regulation by inflammatory cytokines. Front Neurosci. 2014;8:12.
190. Savitz J. The kynurenine pathway: a finger in every pie. Mol Psychiatry. 2020;25:131–47.
191. Sorrells SF, Caso JR, Munhoz CD, Sapolsky RM. The stressed CNS: when glucocorticoids aggravate inflammation. Neuron. 2009;64:33–9.
192. Madrigal JL, Hurtado O, Moro MA, Lizasoain I, Lorenzo P, Castrillo A, Boscá L, Leza JC. The increase in TNF-alpha levels is implicated in NF-kappa B activation and inducible nitric oxide synthase expression in brain cortex after immobilization stress. Neuropsychopharmacology. 2002;26:155–63.
193. Ohgidani M, Kato TA, Sagata N, Hayakawa K, Shimokawa N, Sato-Kasai M, Kanba S. TNF-α from hippocampal microglia induces working memory deficits by acute stress in mice. Brain Behav Immun. 2016;55:17–24.
194. Nguyen KT, Deak T, Will MJ, Hansen MK, Hunsaker BN, Fleshner M, Watkins LR, Maier SF. Time course and corticosterone sensitivity of the brain, pituitary, and serum interleukin-1beta protein response to acute stress. Brain Res. 2000;859:193–201.
195. Fujigaki S, Saito K, Sekikawa K, Tone S, Takikawa O, Fujii H, Wada H, Noma A, Seishima M. Lipopolysaccharide induction of indoleamine 2,3-dioxygenase is mediated dominantly by an IFN-gamma-independent mechanism. Eur J Immunol. 2001;31:2313–8.
196. Tu H, Rady PL, Juelich T, Smith EM, Tyring SK, Hughes TK. Cytokine regulation of tryptophan metabolism in the hypothalamic-pituitary-adrenal (HPA) axis: implications for protective and toxic consequences in neuroendocrine regulation. Cell Mol Neurobiol. 2005;25:673–80.

197. Myint AM, Kim YK. Network beyond IDO in psychiatric disorders: revisiting neurodegeneration hypothesis. Prog Neuro-Psychopharmacol Biol Psychiatry. 2014;48:304–13.
198. Parrott JM, O'Connor JC. Kynurenine 3-monooxygenase: an influential mediator of neuropathology. Front Psych. 2015;6:116.
199. Tavares RG, Tasca CI, Santos CE, Alves LB, Porciúncula LO, Emanuelli T, Souza DO. Quinolinic acid stimulates synaptosomal glutamate release and inhibits glutamate uptake into astrocytes. Neurochem Int. 2002;40:621–7.
200. Guillemin GJ, Smythe G, Takikawa O, Brew BJ. Expression of indoleamine 2,3-dioxygenase and production of quinolinic acid by human microglia, astrocytes, and neurons. Glia. 2005;49:15–23.
201. Pérez-De La Cruz V, Carrillo-Mora P, Santamaría A. Quinolinic acid, an endogenous molecule combining excitotoxicity, oxidative stress and other toxic mechanisms. Int J Tryptophan Res. 2012;5:1–8.
202. Castellano-Gonzalez G, Jacobs KR, Don E, Cole NJ, Adams S, Lim CK, Lovejoy DB, Guillemin GJ. Kynurenine 3-monooxygenase activity in human primary neurons and effect on cellular bioenergetics identifies new neurotoxic mechanisms. Neurotox Res. 2019;35:530–41.
203. Sayre LM, Perry G, Smith MA. Oxidative stress and neurotoxicity. Chem Res Toxicol. 2008;21:172–88.
204. Van der Does AJ. The effects of tryptophan depletion on mood and psychiatric symptoms. J Affect Disord. 2001;64:107–19.
205. Miura H, Ozaki N, Sawada M, Isobe K, Ohta T, Nagatsu T. A link between stress and depression: shifts in the balance between the kynurenine and serotonin pathways of tryptophan metabolism and the etiology and pathophysiology of depression. Stress. 2008;11:198–209.
206. Sublette ME, Postolache TT. Neuroinflammation and depression: the role of indoleamine 2,3-dioxygenase (IDO) as a molecular pathway. Psychosom Med. 2012;74:668–72.
207. La Torre D, Dalile B, de Loor H, Van Oudenhove L, Verbeke K. Changes in kynurenine pathway metabolites after acute psychosocial stress in healthy males: a single-arm pilot study. Stress. 2021;24(6):920–30.
208. Myint K, Jacobs K, Myint AM, Lam SK, Lim YA, Boey CC, Hoe SZ, Guillemin GJ. Psychological stresses in children trigger cytokine- and kynurenine metabolite-mediated abdominal pain and proinflammatory changes. Front Immunol. 2021;12:702301.
209. Vecchiarelli HA, Gandhi CP, Hill MN. Acute psychological stress modulates the expression of enzymes involved in the kynurenine pathway throughout corticolimbic circuits in adult male rats. Neural Plast. 2016;2016:7215684.
210. Strasser B, Becker K, Fuchs D, Gostner JM. Kynurenine pathway metabolism and immune activation: peripheral measurements in psychiatric and co-morbid conditions. Neuropharmacology. 2017;112:286–96.
211. Huang YS, Ogbechi J, Clanchy FI, Williams RO, Stone TW. IDO and kynurenine metabolites in peripheral and CNS disorders. Front Immunol. 2020;11:388.
212. Schwarcz R, Pellicciari R. Manipulation of brain kynurenines: glial targets, neuronal effects, and clinical opportunities. J Pharmacol Exp Ther. 2002;303:1–10.
213. Stone TW, Darlington LG. Endogenous kynurenines as targets for drug discovery and development. Nat Rev Drug Discov. 2002;1:609–20.
214. Miura H, Ando Y, Noda Y, Isobe K, Ozaki N. Long-lasting effects of inescapable-predator stress on brain tryptophan metabolism and the behavior of juvenile mice. Stress. 2011;14:262–72.
215. Dostal CR, Carson Sulzer M, Kelley KW, Freund GG, McCusker RH. Glial and tissue-specific regulation of kynurenine pathway dioxygenases by acute stress of mice. Neurobiol Stress. 2017;7:1–15.

216. Ohta Y, Kubo H, Yashiro K, Ohashi K, Tsuzuki Y, Wada N, Yamamoto Y, Saito K. Effect of water-immersion restraint stress on tryptophan catabolism through the kynurenine pathway in rat tissues. J Physiol Sci. 2017;67:361–72.
217. Pawlak D, Takada Y, Urano T, Takada A. Serotonergic and kynurenic pathways in rats exposed to foot shock. Brain Res Bull. 2000;52:197–205.
218. Coplan JD, George R, Syed SA, Rozenboym AV, Tang JE, Fulton SL, Perera TD. Early life stress and the fate of kynurenine pathway metabolites. Front Hum Neurosci. 2021;15:636144.
219. Fuertig R, Azzinnari D, Bergamini G, Cathomas F, Sigrist H, Seifritz E, Vavassori S, Luippold A, Hengerer B, Ceci A, Pryce CR. Mouse chronic social stress increases blood and brain kynurenine pathway activity and fear behaviour: both effects are reversed by inhibition of indoleamine 2,3-dioxygenase. Brain Behav Immun. 2016;54:59–72.
220. Nold V, Sweatman C, Karabatsiakis A, Böck C, Bretschneider T, Lawless N, Fundel-Clemens K, Kolassa IT, Allers KA. Activation of the kynurenine pathway and mitochondrial respiration to face allostatic load in a double-hit model of stress. Psychoneuroendocrinology. 2019;107:148–59.
221. Margină D, Ungurianu A, Purdel C, Tsoukalas D, Sarandi E, Thanasoula M, Tekos F, Mesnage R, Kouretas D, Tsatsakis A. Chronic inflammation in the context of everyday life: dietary changes as mitigating factors. Int J Environ Res Public Health. 2020;17:4135.
222. Cohen M, Meir T, Klein E, Volpin G, Assaf M, Pollack S. Cytokine levels as potential biomarkers for predicting the development of posttraumatic stress symptoms in casualties of accidents. Int J Psychiatry Med. 2011;42:117–31.
223. Hao Y, Jing H, Bi Q, Zhang J, Qin L, Yang P. Intra-amygdala microinfusion of IL-6 impairs the auditory fear conditioning of rats via JAK/STAT activation. Behav Brain Res. 2014;275:88–95.
224. Jing H, Hao Y, Bi Q, Zhang J, Yang P. Intra-amygdala microinjection of TNF-α impairs the auditory fear conditioning of rats via glutamate toxicity. Neurosci Res. 2015;91:34–40.
225. Rosso IM, Weiner MR, Crowley DJ, Silveri MM, Rauch SL, Jensen JE. Insula and anterior cingulate GABA levels in posttraumatic stress disorder: preliminary findings using magnetic resonance spectroscopy. Depress Anxiety. 2014;31:115–23.
226. Crowley T, Cryan JF, Downer EJ, O'Leary OF. Inhibiting neuroinflammation: the role and therapeutic potential of GABA in neuro-immune interactions. Brain Behav Immun. 2016;54:260–77.
227. Izuno S, Yoshihara K, Sudo N. Role of gut microbiota in the pathophysiology of stress-related disorders: evidence from neuroimaging studies. Ann Nutr Metab. 2021;77:4–10.
228. McVey Neufeld KA, Perez-Burgos A, Mao YK, Bienenstock J, Kunze WA. The gut microbiome restores intrinsic and extrinsic nerve function in germ-free mice accompanied by changes in calbindin. Neurogastroenterol Motil. 2015;27:627–36.
229. Neufeld KM, Kang N, Bienenstock J, Foster JA. Reduced anxiety-like behavior and central neurochemical change in germ-free mice. Neurogastroenterol Motil. 2011;23:255–64.
230. Gareau MG, Wine E, Rodrigues DM, Cho JH, Whary MT, Philpott DJ, Macqueen G, Sherman PM. Bacterial infection causes stress-induced memory dysfunction in mice. Gut. 2011;60:307–17.
231. Bercik P, Denou E, Collins J, Jackson W, Lu J, Jury J, Deng Y, Blennerhassett P, Macri J, McCoy KD, Verdu EF, Collins SM. The intestinal microbiota affect central levels of brain-derived neurotropic factor and behavior in mice. Gastroenterology. 2011;141:599–609.
232. Bercik P, Park AJ, Sinclair D, Khoshdel A, Lu J, Huang X, Deng Y, Blennerhassett PA, Fahnestock M, Moine D, Berger B, Huizinga JD, Kunze W, McLean PG, Bergonzelli GE, Collins SM, Verdu EF. The anxiolytic effect of Bifidobacterium longum NCC3001 involves vagal pathways for gut-brain communication. Neurogastroenterol Motil. 2011;23:1132–9.
233. Bravo JA, Forsythe P, Chew MV, Escaravage E, Savignac HM, Dinan TG, Bienenstock J, Cryan JF. Ingestion of lactobacillus strain regulates emotional behavior and central GABA receptor expression in a mouse via the vagus nerve. Proc Natl Acad Sci U S A. 2011;108:16050–5.

234. Hamer HM, De Preter V, Windey K, Verbeke K. Functional analysis of colonic bacterial metabolism: relevant to health? Am J Physiol Gastrointest Liver Physiol. 2012;302:G1–9.
235. Al-Nedawi K, Mian MF, Hossain N, Karimi K, Mao YK, Forsythe P, Min KK, Stanisz AM, Kunze WA, Bienenstock J. Gut commensal microvesicles reproduce parent bacterial signals to host immune and enteric nervous systems. FASEB J. 2015;29:684–95.
236. Leclercq S, Forsythe P, Bienenstock J. Posttraumatic stress disorder: does the gut microbiome hold the key? Can J Psychiatr. 2016;61:204–13.
237. Breit S, Kupferberg A, Rogler G, Hasler G. Vagus nerve as modulator of the brain-gut axis in psychiatric and inflammatory disorders. Front Psych. 2018;9:44.
238. Jiang H, Ling Z, Zhang Y, Mao H, Ma Z, Yin Y, Wang W, Tang W, Tan Z, Shi J, Li L, Ruan B. Altered fecal microbiota composition in patients with major depressive disorder. Brain Behav Immun. 2015;48:186–94.
239. Szczesniak O, Hestad KA, Hanssen JF, Rudi K. Isovaleric acid in stool correlates with human depression. Nutr Neurosci. 2016;19:279–83.
240. Hemmings SMJ, Malan-Müller S, van den Heuvel LL, Demmitt BA, Stanislawski MA, Smith DG, Bohr AD, Stamper CE, Hyde ER, Morton JT, Marotz CA, Siebler PH, Braspenning M, Van Criekinge W, Hoisington AJ, Brenner LA, Postolache TT, McQueen MB, Krauter KS, Knight R, Seedat S, Lowry CA. The microbiome in posttraumatic stress disorder and trauma-exposed controls: an exploratory study. Psychosom Med. 2017;79:936–46.
241. Jergović M, Bendelja K, Savić Mlakar A, Vojvoda V, Aberle N, Jovanovic T, Rabatić S, Sabioncello A, Vidović A. Circulating levels of hormones, lipids, and immune mediators in post-traumatic stress disorder - a 3-month follow-up study. Front Psych. 2015;6:49.
242. Brenner LA, Forster JE, Stearns-Yoder KA, Stamper CE, Hoisington AJ, Brostow DP, Mealer M, Wortzel HS, Postolache TT, Lowry CA. Evaluation of an immunomodulatory probiotic intervention for veterans with co-occurring mild traumatic brain injury and posttraumatic stress disorder: a pilot study. Front Neurol. 2020;11:1015.
243. Zhutovsky P, Thomas RM, Olff M, van Rooij SJH, Kennis M, van Wingen GA, Geuze E. Individual prediction of psychotherapy outcome in posttraumatic stress disorder using neuroimaging data. Transl Psychiatry. 2019;9:326.
244. Li F, Xiang H, Lu J, Chen Z, Huang C, Yuan X. Lycopene ameliorates PTSD-like behaviors in mice and rebalances the neuroinflammatory response and oxidative stress in the brain. Physiol Behav. 2020;224:113026.
245. Himmerich H, Willmund GD, Zimmermann P, Wolf JE, Bühler AH, Kirkby KC, Dalton B, Holdt LM, Teupser D, Wesemann U. Serum concentrations of TNF-α and its soluble receptors during psychotherapy in German soldiers suffering from combat-related PTSD. Psychiatr Danub. 2016;28:293–8.
246. Tucker P, Ruwe WD, Masters B, Parker DE, Hossain A, Trautman RP, Wyatt DB. Neuroimmune and cortisol changes in selective serotonin reuptake inhibitor and placebo treatment of chronic posttraumatic stress disorder. Biol Psychiatry. 2004;56:121–8.

# Sleep Immune Cross Talk and Insomnia

12

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**Abstract**

Sleep and immunity have bidirectional relationships. In this chapter, we review the links between sleep and immunity, focusing on immune changes occurring in the insomnia disorder. During physiological sleep, there is a decrease of pro-inflammatory cytokines (IL-1, IL-6 and TNF-α) and a decrease of anti-inflammatory cytokines (IL-4, IL-10). Examinations of ratios of pro-inflammatory and anti-inflammatory cytokines allow to identify rather a pro-inflammatory activity at the beginning of the night and confirm then anti-inflammatory during the second part of the night. Immune cells, as NK-cells, decrease in the blood, due to their migration to secondary lymphoid organs, but

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their activity increases. Inversely, a short sleep duration appears associated with increased inflammatory processes and increased risk of infection.

Only few studies have investigated changes in immunity in patients with insomnia disorder. These studies suggest that insomnia disorder is related to deregulation of the immune system, with an increase in the level of pro-inflammatory cytokines and change in rate of secretion and a decrease in the level of lymphocyte. Insomnia treatments, particularly cognitive behavioral therapy (CBT-I), seems to have a restorative effect not only on sleep, but also on the associated inflammation. Melatonin also seems to reduce inflammation in patients suffering from insomnia disorder.

More studies are necessary to better understand the pathophysiology of changes in immune system in patients suffering from insomnia disorders and their clinical implications.

**Keywords**

Insomnia · Immunity · Inflammation · Sleep immune cross talk · Sleep disorders · Sleep deprivation · Sleep loss

## 12.1 Introduction

Sleep and immunity have bidirectional relationships and activation of the immune system can affect sleep, and similarly, sleep has an effect on the immune system [1–3].

At the end of the nineteenth century, some experimental studies conducted in animals have shown that particularly total sleep deprivation may led to lethal consequences several or 15 days after [4, 5]. One hypothesis was that these animals died as a result of bacteremia caused by sleep deprivation [6]. These observations suggested an important role of sleep on immunity, which indeed remained to be clarified in humans.

Among causes, which may lead to sleep deprivation, insomnia may play a role, and it is one of the most frequent sleep disorders in the general population. Insomnia is defined by the ICSD-3 (International Classification of Sleep Disorders version 3) [7] as at least one sleep disorder (difficulty in falling asleep, difficulty in maintaining sleep, waking up too early, sleep time less than 6 h). These symptoms must occur with a specific frequency (at least 3 times a week) and persist for a certain time (for at least 3 months), in an adequate night sleep context, with repercussions on daytime functioning (fatigue, irritability, concentration and memory problems, mood disorders, alteration of social life, etc.). The prevalence of insomnia in the different studies varies due to the great heterogeneity of the definitions, populations, and methodologies used [8]. Nevertheless, about one-third of the general population reports occasional difficulty falling or staying asleep during the night, and for approximately 6–13% of the population, these difficulties are experienced more regularly and result in distress and negative consequences for daytime functioning

[9]. The rigorous application of diagnostic criteria for insomnia disorder places its prevalence at around 10% in the general population [10]. Insomnia is a severe disorder leading to medical and neurodegenerative disorders, psychiatric, addictive disorders, and increased suicide risk, all conditions potentially associated with insomnia-related inflammation [11–14].

This work will review the links between sleep and immunity, starting by outlining relationships between the immune system and physiological sleep. In a second part, we will focus on immune changes occurring in the insomnia disorder.

## 12.2 Changes in Immunity During Physiological Sleep

### 12.2.1 Cytokines

#### 12.2.1.1 Pro-inflammatory Cytokines (IL-1, IL-6, TNF-α)

Several studies have found that pro-inflammatory cytokines (IL-1, IL-6, and TNF-α) decrease during normal sleep, suggesting an anti-inflammatory function of sleep. In contrast, during sleep deprivation, an increase in blood levels of pro-inflammatory cytokines is observed [15].

IL-2 is a cytokine that mediates adaptive immunity. Under normal circumstances, no changes are observed during sleep. However, after vaccination, an increase in IL-2 levels has been observed, which is not found in case of prolonged wakefulness after vaccination [15].

#### 12.2.1.2 Anti-inflammatory Cytokines (IL-4, IL-10)

Conversely, an decrease in IL-4 and IL-10 anti-inflammatory cytokines levels have been reported during sleep [15].

#### 12.2.1.3 Ratio of Pro- and Anti-inflammatory Cytokines

Dimitrov et al. [16] found an increase in the TNF-α/IL-4 ratio during the first part of sleep (thus in favor of a pro-inflammatory activity), which reverses in the second part of the night (in favor of an anti-inflammatory activity). In line with these observations, Axelsson et al. [17] reported an increase in the IL-2/IL-4 ratio in the case of prolonged sleep deprivation (pro-inflammatory activity).

Taken together these findings suggest a decrease in anti-inflammatory activity during sleep when measuring cytokine levels. Examinations of ratios of pro-inflammatory and anti-inflammatory cytokines allow to identify rather a pro-inflammatory activity at the beginning of the night and confirm then anti-inflammatory during the second part of the night.

### 12.2.2 Immunity Cells

In 1997, Born et al. found a decrease in the blood level of leukocytes and Natural Killers cells (NK-cells) during sleep [18] and hypothesized that this decrease is

related to a redistribution of immune cells to the lymph nodes and organs during sleep. Ruiz et al. [19] validate this hypothesis in skin transplanted from mice, showing a redistribution of immune cells during sleep to the spleen and lymph nodes, in contrast to sleep deprived mice.

Irwin et al. [20] show that NK-cells activity actually increases during sleep and decreases during sleep deprivation. Moreover, if sleep deprivation is prolonged, a rebound in NK cell activity is observed. This observation is identical for lymphocytes and monocytes [20]. Recently, Ruiz et al. [21] reported that a total of two nights of sleep deprivation resulted in increased leukocytes and neutrophils in healthy men compared to their baseline values. After 24 h of sleep recovery, those figures returned to the values observed at baseline.

In summary, during sleep, immune cells decrease in the blood, due to their migration to secondary lymphoid organs. In addition to this migration, the activity of immunocompetent cells increases during sleep.

### 12.2.3 Effects of Sleep on Adaptive Immunity: The Example of Vaccination

Vaccination is an example of adaptive immunity. The first observation was conducted by Spiegel et al. [22], who observed the effects of sleep deprivation on the creation of antibodies specific to the H1N1 flu virus after vaccination. A first group of patients received the H1N1-vaccine and then underwent sleep deprivation (4 h of sleep per night for 6 nights) and was compared with a second group that also received the vaccine but without sleep deprivation. Patients without sleep deprivation had 2.5 times higher levels of H1N1-specific antibodies than those who were sleep deprived. This study suggests that sleep has a key role and allow the immune system to develop antibodies. Later studies confirmed that a single night of sleep deprivation resulted in decreased antibody levels following vaccination against hepatitis A and B [23].

If decreased sleep duration prevents the humoral response following vaccination, it may be so hypothesized that increased sleep duration may enhance the adaptive immunity response following vaccination. Prathner et al. [24] compared the humoral response following hepatitis B vaccination in individuals sleeping less than 6 h and more than 7 h and found that the proportion of individuals achieving effective protection was 3.5 times higher in the longer sleep group.

Just as sleep promotes memory consolidation at the brain level, it seems to promote immune memory and would allow for better adaptive immunity (Fig. 12.1).

### 12.2.4 Sleep Response to Acute Immune Activation

#### 12.2.4.1 Sleep Architecture Modifications during Infection

During acute infection, increased sleep is considered as a host defense response. Indeed, during acute infection in general, non-rapid eye movement sleep

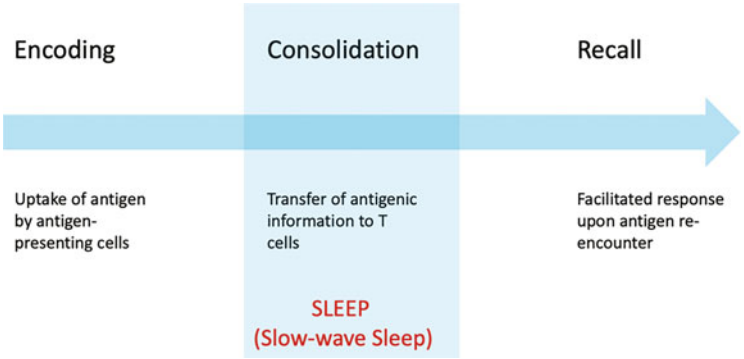


**Fig. 12.1** Effects of sleep on immune memory. (Adapted from Besedovsky, 2019 [23])

(NREM-Sleep) and more specifically slow wave sleep (SWS) are increased, together with febrile response [25, 26]. On the contrary, rapid eye movement sleep (REM sleep) is rather diminished or suppressed. This sleep response to infection is mediated by cytokines. Indeed, pro-inflammatory cytokines, such as IL-1 and TNF-α, are described as NREM-sleep inducers with fever; however, IL-6 induces fever but impairs sleep (sleep fragmentation). Overall, animal studies suggest that most pro-inflammatory cytokines are NREM sleep promoting, while anti-inflammatory cytokines are NREM sleep reducing [23].

## 12.3 Insomnia and Immunity

### 12.3.1 Effects of a Reduced Sleep Duration on Immunity

There seems to be a relationship between the duration of sleep and immunity (cellular and humoral). Several studies in humans have shown that short habitual sleep duration, i.e., less than 5–6 hours, increases cardiovascular and mental disorders and increases the mortality risk [27–29]. Short sleep duration has been showed to be related to cardiovascular diseases, metabolic diseases, some forms of cancers, and an neuropsychiatric diseases [1], which all involve immune dysregulations.

A study conducted in 2500 elderly people, who were followed for 7 years, found that a sleep duration of less than 5 h was associated with an increase in proinflammatory cytokines (IL-6, TNF-α) and CRP and was associated with an increased mortality [30]. Another prospective study conducted in 3000 elderly subjects, who were followed for 9 years, found an association between pro-inflammatory cytokines (IL-6, TNF-α), CRP, reduced sleep duration, and mortality [31]. In line with this, in adolescents sleeping less than 8 h per night was associated with an increase in leukocytes, monocytes, neutrophils, and T lymphocytes [32].

Several studies have found an increased risk of developing an infection in people with shortened sleep duration. Indeed, Patel et al. [33] found an increased risk of developing a lung infection in people sleeping less than 5 h per night. Another study found that people who reported sleeping less than 7 h per night were more likely to develop respiratory infections than people who reported sleeping more than 8 h per night [34].

So shorter sleep durations appear associated with increased inflammatory processes and increased risk of infections.

### 12.3.2 Insomnia and Immunity

A study conducted by Donners et al. [35] in German students found that students who reported being sick frequently reported more insomnia symptoms (on the SLEEP-50 questionnaire) than students who reported being rarely sick.

Few studies have investigated changes in immunity in patients with insomnia disorder (Table 12.1). A study reported that chronic insomnia has been shown to be associated with increased secretion of TNF-α and IL-6 during the day, combined with hypersecretion of cortisol, an arousal hormone that leads to daytime fatigue, sleepiness, and poor sleep quality [36]. In this study, authors observed in individuals with insomnia, compared to controls, a shift of the peak levels of systemic IL-6 and TNF-α from night time to daytime. So the authors hypothesized that these daytime increases in inflammatory cytokines may explain increased fatigue experienced during the day. Another interesting study reported increased levels of IL-6 during the night compared to day time, in 11 patients suffering from insomnia disorder [37]. Moreover, in the same study, longer PSG-derived nocturnal wake duration was related to higher IL-6 levels. Consistent results were found in the study by Floam

**Table 12.1** Summary of changes in immunity during physiological sleep, sleep deprivation, and insomnia disorder

| | Physiological sleep | Sleep deprivation | Insomnia disorder |
|---|---|---|---|
| *Leucocytes* | | | |
| Lymphocytes | ↓ | ↑ | ? |
| Monocytes | ↓ | ↑ | ? |
| NK-cells | | | |
| · Count<br>· Activity | ↓<br>↑ | ↑<br>↓ | =<br>= |
| *Cytokines* | | | |
| Pro-inflammatory IL-1, IL-6, TNF-α | ↓ | ↑ | ↑ |
| Anti-inflammatory (IL-4, IL-10) | ↓ | = | ? |
| *CRP* | | | |
| CRP | = | ↑ | ↑ |

et al. [38], who found a higher inflammatory score (composite score based on IL-6, CRP and monocyte levels) in 29 patients with insomnia disorder, compared with good sleepers. This increase in inflammation was associated with an objective decrease in sleep duration (measured by actigraphy) of 45 min in patients with insomnia.

As mentioned above, sleep plays an important role in the consolidation of adaptive immunity and humoral memory. A study that compared 133 healthy college students with or without insomnia disorder (based on DSM-5 criteria) found that students who reported insomnia had a poorer humoral response (lower antibody levels) to influenza vaccination than students with no sleep problems [39]. In a another study, Savard et al. [40] compared the count of lymphocytes in patients with chronic insomnia (based on DSM-IV criteria) and in patients with no sleep disorders. The authors found a lower count of lymphocyte subpopulation (i.e., total T cells, CD4 T cells, and CD8 T cells) in patients with chronic insomnia than in healthy controls. They did not find any differences regarding total leukocyte counts, NK-cell activity, or production of IL-1, IL-2, and IFN-γ. However, in this study, individuals with insomnia and healthy controls did not differ on PSG-sleep parameters, while the group with a diagnosis of insomnia reported subjectively a longer nocturnal wake duration and lower sleep efficiency. This lack of differences in objective sleep parameters on PSG may have explained the absence of difference in the total leukocyte count in both populations. Only one study [41] explored telomere length in insomnia disorder and found that the presence of insomnia may accelerate cellular aging in the later years of life (>70 years).

The results of these different studies suggest that the insomnia disorder is related to deregulations of the immune system: increase in the level of pro-inflammatory cytokines and change in the rate of secretion and decrease in the level of lymphocytes.

### 12.3.3 Insomnia Treatments and Immunity

According to international guidelines, insomnia treatment includes non-pharmacological sleep intervention such as cognitive behavioral therapy (CBT-I) and pharmacological options such as GABAa-receptor allostatic modulators and melatonin receptors agonists, among antidepressants doxepine and dual orexin receptor antagonists (DORAs).

#### 12.3.3.1 Non-pharmacological Sleep Intervention: Cognitive Behavioral Therapy (CBT-I)

The gold standard treatment for chronic insomnia is cognitive behavioral therapy [42]. Irwin et al. [43] found that the use of CBT-I in 100 patients suffering from chronic insomnia was accompanied not only by clinical improvement but also by a decrease in the level of pro-inflammatory cytokines (IL-6, TNF-α) and CRP. This decrease persisted at 2 months of follow-up for cytokines, and up to 16 months of follow-up for CRP. Several other studies have found this decrease in CRP after

CBT-I in insomniacs, as well as in IL-1 and IL-18 levels [23]. A study using genome-wide transcriptional profiling found that the transcription of genes involved in inflammation were downregulated after treatment with CBT-I [43]. Transcription of genes involved in IFN and antibody responses were upregulated [43].

#### 12.3.3.2 Pharmacological Sleep Intervention

##### GABAa-Receptor Allostatic Modulators

Hypnotics and anxiolytics of the BZD class improve sleep efficiency by reducing the latency of sleep onset and arousals during the night [44]. We have not found any specific study on the effect of Z-drugs and benzodiazepines on immunity in humans. We found two studies conducted in animals and involving benzodiazepines: In the first study, mice were treated with midazolam daily after a burn injury; the authors observed a decrease in IL-1, TNF-**α**, IL-6, IL-10, and TGF-b levels compared to saline-treated mice [45]. In a second study, using the same treatment with midazolam, investigated whether psychological stress could alter survival following *Pseudomonas aeruginosa* infection. The results showed that midazolam had a protective effect in mice [46].

Together, these two studies suggest that benzodiazepines can modulate the immune system and inflammatory mediators. Another study hypothesizes that the use of benzodiazepines may help healing in cases of skin infection, through their indirect effect on the immune system, mediated by their effect on sleep [44].

##### Orexin Receptor Antagonists (DORAs)

We have not found any specific study on the effect of DORAs on immunity in humans in relation to insomnia. Indeed, a study was conducted in patients with delirium to examine the efficacy of suvorexant, as a therapeutic agent for the treatment of delirium and C-reactive protein levels. Suvorexant exhibited to decrease levels of C-reactive protein, suggesting an anti-inflammatory effect [47].

#### 12.3.3.3 Antidepressants

The only antidepressant suggested for insomnia treatment is doxepine. At low doses, it is very high selective for H1 receptors and can produce selective H1 blockade. It binds with histamine receptors for 100 times more than that of norepinephrine and serotonin receptors, and this inhibits the arousal pathway.

We have not found any specific study on the effect of doxepine on immunity in humans. A study investigated the effects of doxepin on levels of tumor necrosis factor-alpha (TNF-α) in the rat hippocampus following repeated restraint stress. A study has shown that TNF-α level was increased significantly in stress group and low dose of doxepin decreased TNF-α level [48].

#### 12.3.3.4 Melatonin Receptor Agonists

The use of melatonin in the treatment of insomnia seems to have an effect on the decrease in CRP levels. In a meta-analysis, Zareradeh et al. [49] show that the use of melatonin as a dietary supplement, at a dose of 3–25 mg/day for several months, is

accompanied by a decrease in the levels of pro-inflammatory cytokines (IL-6, TNF-α) and CRP. The authors conclude that melatonin is useful in reducing low-grade inflammation. In another study, Shimizu et al. [50] found an immunomodulatory, anti-inflammatory and antioxidant effect of Ramelteon in subjects suffering from a chronic insomnia disorder. Melatonin has an inhibitory role on pro-inflammatory cytokines and prostaglandins. It also seems to have a role in T-cell proliferation.

## 12.4 Conclusion

A bidirectional link seems to exist between the immune system and sleep. Few studies have looked at changes in immunity in insomnia. The few studies found tend to show an increase in the level of pro-inflammatory cytokines in insomnia and a decrease in lymphocytes. It is interesting to note that the various insomnia treatments, in particular CBT-I, have a clinical impact on insomnia, but also on the regulation of immunity.

Further studies are needed to better understand the links between insomnia and the immune system and their clinical consequences.

## References

1. Irwin MR. Why sleep is important for health: a psychoneuroimmunology perspective. Annu Rev Psychol. 2015;66:143–72.
2. Irwin MR, Olmstead R, Carroll JE. Sleep disturbance, sleep duration, and inflammation: a systematic review and meta-analysis of cohort studies and experimental sleep deprivation. Biol Psychiatry. 2016;80(1):40–52.
3. Irwin MR, Opp MR. Sleep health: reciprocal regulation of sleep and innate immunity. Neuropsychopharmacology. 2017;42(1):129–55.
4. Bentivoglio M, Grassi-Zucconi G. The pioneering experimental studies on sleep deprivation. Sleep. 1997;20(7):570–6.
5. Rechtschaffen A, Gilliland MA, Bergmann BM, Winter JB. Physiological correlates of prolonged sleep deprivation in rats. Science. 1983;221(4606):182–4.
6. Everson CA, Toth LA. Systemic bacterial invasion induced by sleep deprivation. Am J Physiol Regul Integr Comp Physiol. 2000;278(4):R905–16.
7. Sateia MJ. International classification of sleep disorders-third edition: highlights and modifications. Chest. 2014;146(5):1387–94.
8. Ohayon MM. Epidemiology of insomnia: what we know and what we still need to learn. Sleep Med Rev. 2002;6(2):97–111.
9. Morin CM, LeBlanc M, Bélanger L, Ivers H, Mérette C, Savard J. Prevalence of insomnia and its treatment in Canada. Can J Psychiatry. 2011;56(9):540–8.
10. Elsevier, N A-C. Insomnie chez l'adulte [Internet]. Elsevier Connect. [cité 1 nov 2021]. https://www.elsevier.com/fr-fr/connect/medecine/insomnie-chez-ladulte
11. Baglioni C, Battagliese G, Feige B, Spiegelhalder K, Nissen C, Voderholzer U, et al. Insomnia as a predictor of depression: a meta-analytic evaluation of longitudinal epidemiological studies. J Affect Disord. 2011;135(1–3):10–9.
12. Geoffroy PA, Oquendo MA, Courtet P, Blanco C, Olfson M, Peyre H, et al. Sleep complaints are associated with increased suicide risk independently of psychiatric disorders: results from a national 3-year prospective study. Mol Psychiatry. 2021;26(6):2126–36.

13. Hertenstein E, Feige B, Gmeiner T, Kienzler C, Spiegelhalder K, Johann A, et al. Insomnia as a predictor of mental disorders: a systematic review and meta-analysis. Sleep Med Rev. 2019;43:96–105.
14. Morin CM, Drake CL, Harvey AG, Krystal AD, Manber R, Riemann D, et al. Insomnia disorder. Nat Rev Dis Primers. 2015;1:15026.
15. Poluektov MG. Sleep and immunity. Neurosci Behav Physiol. 2021;51(5):609–15.
16. Dimitrov S, Lange T, Tieken S, Fehm HL, Born J. Sleep associated regulation of T helper 1/T helper 2 cytokine balance in humans. Brain Behav Immun. 2004;18(4):341–8.
17. Axelsson J, Rehman J, Akerstedt T, Ekman R, Miller GE, Höglund CO, et al. Effects of sustained sleep restriction on mitogen-stimulated cytokines, chemokines and T helper 1/T helper 2 balance in humans. PLoS One. 2013;8(12):e82291.
18. Born J, Lange T, Hansen K, Mölle M, Fehm HL. Effects of sleep and circadian rhythm on human circulating immune cells. J Immunol. 1997;158(9):4454–64.
19. Ruiz FS, Andersen ML, Guindalini C, Araujo LP, Lopes JD, Tufik S. Sleep influences the immune response and the rejection process alters sleep pattern: evidence from a skin allograft model in mice. Brain Behav Immun. 2017;61:274–88.
20. Irwin M, McClintick J, Costlow C, Fortner M, White J, Gillin JC. Partial night sleep deprivation reduces natural killer and cellular immune responses in humans. FASEB J. 1996;10(5):643–53.
21. Ruiz FS, Andersen ML, Martins RC, Zager A, Lopes JD, Tufik S. Immune alterations after selective rapid eye movement or total sleep deprivation in healthy male volunteers. Innate Immun. 2012;18(1):44–54.
22. Spiegel K, Sheridan JF, Van Cauter E. Effect of sleep deprivation on response to Immunizaton. JAMA. 2002;288(12):1471–2.
23. Besedovsky L, Lange T, Haack M. The sleep-immune crosstalk in health and disease. Physiol Rev. 2019;99(3):1325–80.
24. Prather AA, Hall M, Fury JM, Ross DC, Muldoon MF, Cohen S, et al. Sleep and antibody response to Hepatitis B vaccination. Sleep. 2012;35(8):1063–9. https://doi.org/10.5665/sleep.1990.
25. Krueger JM, Majde JA. Sleep as a host defense: its regulation by microbial products and cytokines. Clin Immunol Immunopathol. 1990;57(2):188–99.
26. Benca R, Quintas J. Sleep and host defenses: a review. Sleep. 1997;20(11):1027–37.
27. Cappuccio FP, D'Elia L, Strazzullo P, Miller MA. Sleep duration and all-cause mortality: a systematic review and meta-analysis of prospective studies. Sleep. 2010;33(5):585–92.
28. Yin J, Jin X, Shan Z, Li S, Huang H, Li P, et al. Relationship of sleep duration with all-cause mortality and cardiovascular events: a systematic review and dose-response meta-analysis of prospective cohort studies. J Am Heart Assoc. 2017;6(9):e005947.
29. Geoffroy PA, Tebeka S, Blanco C, Dubertret C, Le Strat Y. Shorter and longer durations of sleep are associated with an increased twelve-month prevalence of psychiatric and substance use disorders: findings from a nationally representative survey of US adults (NESARC-III). J Psychiatr Res. 2020;124:34–41.
30. Smagula SF, Stone KL, Redline S, Ancoli-Israel S, Barrett-Connor E, Lane NE, et al. Actigraphy- and polysomnography-measured sleep disturbances, inflammation, and mortality among older men. Psychosom Med. 2016;78(6):686–96.
31. Hall MH, Smagula SF, Boudreau RM, Ayonayon HN, Goldman SE, Harris TB, et al. Association between sleep duration and mortality is mediated by markers of inflammation and health in older adults: the health, aging and body composition study. Sleep. 2015;38(2):189–95.
32. Pérez de Heredia F, Garaulet M, Gómez-Martínez S, Díaz LE, Wärnberg J, Androutsos O, et al. Self-reported sleep duration, white blood cell counts and cytokine profiles in European adolescents: the HELENA study. Sleep Med. 2014;15(10):1251–8.
33. Patel SR, Malhotra A, Gao X, Hu FB, Neuman MI, Fawzi WW. A prospective study of sleep duration and pneumonia risk in women. Sleep. 2012;35(1):97–101.
34. Cohen S, Doyle WJ, Alper CM, Janicki-Deverts D, Turner RB. Sleep habits and susceptibility to the common cold. Arch Intern Med. 2009;169(1):62–7.

35. Donners AAMT, Tromp MDP, Garssen J, Roth T, Verster JC. Perceived immune status and sleep: a survey among Dutch students. Sleep Disord. 2015;2015:1–5.
36. Vgontzas AN, Zoumakis M, Papanicolaou DA, Bixler EO, Prolo P, Lin H-M, et al. Chronic insomnia is associated with a shift of interleukin-6 and tumor necrosis factor secretion from nighttime to daytime. Metabolism. 2002;51(7):887–92.
37. Burgos I, Richter L, Klein T, Fiebich B, Feige B, Lieb K, et al. Increased nocturnal interleukin-6 excretion in patients with primary insomnia: a pilot study. Brain Behav Immun. 2006;20(3): 246–53.
38. Floam S, Simpson N, Nemeth E, Scott-Sutherland J, Gautam S, Haack M. Sleep characteristics as predictor variables of stress systems markers in insomnia disorder. J Sleep Res. 2015;24(3): 296–304.
39. Taylor DJ, Kelly K, Kohut ML, Song K-S. Is insomnia a risk factor for decreased influenza vaccine response? Behav Sleep Med. 2017;15(4):270–87.
40. Savard J, Laroche L, Simard S, Ivers H, Morin CM. Chronic insomnia and immune functioning: psychosomatic medicine. Marsyas. 2003;65(2):211–21.
41. Carroll JE, Irwin MR, Levine M, Seeman TE, Absher D, Assimes T, et al. Epigenetic aging and immune senescence in women with insomnia symptoms: findings from the Women's health initiative study. Biol Psychiatry. 2017;81(2):136–44.
42. van Straten A, van der Zweerde T, Kleiboer A, Cuijpers P, Morin CM, Lancee J. Cognitive and behavioral therapies in the treatment of insomnia: a meta-analysis. Sleep Med Rev. 2018;38:3–16.
43. Irwin MR, Olmstead R, Carrillo C, Sadeghi N, Breen EC, Witarama T, et al. Cognitive behavioral therapy vs. Tai Chi for late life insomnia and inflammatory risk: a randomized controlled comparative efficacy trial. Sleep. 2014;37(9):1543–52.
44. Egydio F, Pires G, Tufik S, Andersen M. Wound-healing and benzodiazepines: does sleep play a role in this relationship? Clinics. 2012;67(7):827–30.
45. Babcock GF, Hernandez L, Yadav E, Schwemberger S, Dugan A. The burn wound inflammatory response is influenced by midazolam. Inflammation. 2012;35(1):259–70.
46. Dugan AL, Gregerson KA, Neely A, Gardner J, Noel GJ, Babcock GF, et al. Mice treated with a benzodiazepine had an improved survival rate following Pseudomonas aeruginosa infection. J Burn Care Res. 2010;31(1):1–12.
47. Okino K, Yamada H, Tomioka H, Nozaki S, Iwanami A, Inamoto A. Use of Suvorexant and antipsychotics in the treatment of delirium after infectious diseases: a retrospective study. J Clin Psychopharmacol. 2021;41:589–93.
48. Azadbakht AA, Radahmadi M, Javanmard SH, Reisi P. The effects of doxepin on stress-induced learning, memory impairments, and TNF-α level in the rat hippocampus. Res Pharm Sci. 2015;10(5):460–5.
49. Zarezadeh M, Khorshidi M, Emami M, Janmohammadi P, Kord-varkaneh H, Mousavi SM, et al. Melatonin supplementation and pro-inflammatory mediators: a systematic review and meta-analysis of clinical trials. Eur J Nutr. 2020;59(5):1803–13.
50. Shimizu N, Nozawa M, Sugimoto K, Yamamoto Y, Minami T, Hayashi T, et al. Therapeutic efficacy and anti-inflammatory effect of ramelteon in patients with insomnia associated with lower urinary tract symptoms. Res Rep Urol. 2013;5:113–9.

# Obsessive-Compulsive Disorder, PANDAS, and Tourette Syndrome: Immuno-inflammatory Disorders

13

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**Abstract**

In the last years, much focus has been given to the possible role of inflammatory and immunologic alterations in the pathophysiology of obsessive-compulsive disorder (OCD) and some related conditions, such as pediatric autoimmune neuropsychiatric disorders associated with streptococcal infection (PANDAS) and Tourette syndrome (TS). Although the matter is intriguing, the available data are still controversial and/or limited. Therefore, the aim of this chapter was at reviewing and commenting on the literature on possible dysfunctions of inflammatory and immune system processes in OCD, PANDAS, and TS.

This narrative review was carried out through searching PubMed and Google Scholar for English language papers from January 1985 to December 31, 2021.

The data gathered up to now would suggest that the mechanisms involved might be heterogeneous according to the age of the patients and the disorder examined. Indeed, PANDAS seem more related to infections triggering autoimmunity not necessarily following group A beta-hemolytic streptococcal (GABHS) infection, as supposed in the past. Autoimmunity seems also important

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in TS, if coupled with an individual vulnerability that can be genetic and/or environmental. The data in adult OCD, albeit scattered and sometimes obtained in small samples of patients, would indicate that immune system and inflammatory processes are involved in the pathophysiology of the disorder. However, it is still unclear to conclude whether they are primary or secondary phenomena.

In conclusion, taken together, the current findings pave that way towards novel and promising domains to explore the pathophysiology of OCD and related disorders, as well towards the development of innovative therapeutic strategy beyond current pharmacological paradigms.

**Keywords**

OCD · PANDAS · Tourette syndrome · Neuro-inflammation · Immune system · Cytokines · Neuropsychiatric disorders · Childhood

## Abbreviations

| | |
|---|---|
| 5-HT | 5-Hydroxytryptamine |
| ABGA | Anti-basal ganglia antibodies |
| BBB | Blood-brain barrier |
| BDNF | Brain-derived neurotrophic factor |
| CASPR2 | Contactin-associated protein-like 2 |
| CNS | Central nervous system |
| COX-2 | Ciclooxigenase-2 |
| CSF | Cerebrospinal fluid |
| CSTC | Cortico-striatal-thalamo-cortical |
| DSM-5 | Fifth edition of the Diagnostic and Statistical Manual of Mental Disorders |
| GABA | Gamma-aminobutyric acid |
| GABARAP | GABA receptor-associated protein |
| GABHS | Group A beta-hemolytic streptococcus |
| HCs | Healthy controls |
| HPA | Hypothalamic-pituitary axis |
| IDO | Indoleamine-2,3-dehydrogenase |
| IFN | Interferon |
| IL | Interleukin |
| LPS | Lipopolysaccharide |
| MS | Multiple sclerosis |
| NK | Natural killer |
| NSAIDs | Nonsteroidal anti-inflammatory drugs |
| OCD | Obsessive-compulsive disorder |
| OCRDs | Obsessive-compulsive and related disorders |
| OCS | Obsessive-compulsive symptoms |

| | |
|---|---|
| PANDAS | Pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections |
| PANS | Pediatric acute-onset neuropsychiatric syndrome |
| PD | Parkinson's disease |
| PET | Positron emission tomography |
| ROS | Reactive oxygen species |
| SLE | Systemic lupus erythematosus |
| sTNFR1 | Soluble TNF receptor-1 |
| sTNFR2 | Soluble TNF receptor-2 |
| Th | T helper |
| TM | Transverse myelitis |
| TNF | Tumor necrosis factor |
| Treg | T regulatory |
| TS | Tourette syndrome |
| VT | Distribution volume |
| Y-BOCS | Yale-Brown obsessive-compulsive scale |

## 13.1 Introduction

The study of the complex interactions between the nervous and the immune systems represents one of the most intriguing fields of research in recent years [1]. Not surprisingly, the immune system has been considered to play an important role in the pathophysiology of several neuropsychiatric disorders [2], such as Alzheimer's disease (AD), Parkinson's disease (PD), HIV encephalopathy, multiple sclerosis, transverse myelitis, dementia, schizophrenia (SZ), depression, panic disorder, social phobia, post-traumatic stress disorder, and obsessive-compulsive disorder (OCD) [1, 3–5]. Besides OCD, a role of inflammation has been also suggested in related conditions as pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections (PANDAS) and Tourette syndrome (TS), based on well documented findings collected from both animal and human studies.

Obsessive-compulsive disorder is a common psychiatric disorder with a prevalence of about 2.5%, similarly distributed in both genders in adulthood, although disagreement does exist [6]. It is characterized by obsessions and/or compulsions. Obsessions are recurrent, persistent, intrusive, and unwanted thoughts, urges, or images that cause marked anxiety or distress. The individual tries to ignore or suppress such thoughts, urges, or images or to neutralize them by performing a compulsion that is a repetitive behavior or mental act. The aim of compulsions is to prevent or reduce anxiety or distress, and some dreaded events or situations, although compulsions are not connected in a realistic way to what they are supposed to prevent or neutralize, or are clearly excessive [7]. The evidence that obsessive-compulsive symptoms (OCS) are present even in several other disorders has led to the conceptualization of the new chapter of the fifth edition of the *Diagnostic and*

*Statistical Manual of Mental Disorders (DSM-5)* [7] called "obsessive-compulsive and related disorders" (OCRDs) that, besides OCD, include body dysmorphic disorder, hoarding disorder, trichotillomania (hair-pulling disorder), excoriation (skin-picking disorder), and other.

Despite the impressive achievements obtained in the treatment of OCD since the 1980s of the last century, a large percentage of OCD subjects still show unsatisfactory response to first-line treatments [8, 9] mainly targeting the serotonin (5-hydroxytryptamine, 5-HT). Therefore, other neurotransmitters, such as dopamine and glutamate [10–12], and systems, in particular the immune one, have been proposed [13–18].

Different immunologic processes have been highlighted in both OCD and OCRDs, with more consistent data gathered in children than in adult patients [15, 17, 19]. Pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections (PANDAS) represent a childhood-onset clinical entity characterized by the sudden onset of OCD, tics, and other behavioral symptoms, with a temporal relationship with group A beta-hemolytic streptococcal (GABHS) infection that has become the paradigm of an autoimmune model for OCD [20–23]. Tourette syndrome (TS) is a neuropsychiatric disorder, usually with childhood onset, characterized by the presence of multiple motor tics and one or more vocal tics [24] and representing, once more, a clinical entity in which immune system dysregulations seem to play a pivotal role [25, 26]. On the contrary, data in adult OCD patients are more heterogeneous and scattered. A wide range of mediators and processes of the immune system are supposed to be involved, such as cytokines, microglia cells, genetic and fetal-maternal immune interactions, and anti-basal ganglia antibodies (ABGA) [27–32].

Given the potential of this topic in the perspective of both pathophysiology and novel treatment options, the aim of this chapter was to review the current literature on the relationships between the immune system and OCD, PANDAS, and TS.

## 13.2 Methods

The narrative review was carried out through searching PubMed and Google Scholar for English language papers from January 1985 to December 31, 2021. The keywords used and combined with "OCD" or "OCD symptoms" were "childhood" or "adulthood" or "pathophysiology," or "immune system," "inflammation," or "neuroinflammation," or "cytokines" or "streptococcus infections" or "PANDAS" or "PANS" or "CANS," or "Tourette syndrome," or "antiobsessional drugs." Articles were searched by all authors: they agreed not to include conference abstracts or posters, while case reports were considered if published in indexed journals. The following inclusion criteria were adopted: studies carried out in clinical samples of children/adolescents and/or adults; reliable diagnosis of OCD according to structured interviews and standardized criteria; and use of reliable laboratory tests for biological measures. At the end, a total of 183 papers were included in the review.

## 13.3 Obsessive-Compulsive Disorder

Currently, the immuno-inflammatory hypothesis of OCD can be considered one of the most fascinating and promising research domains [12, 17, 33, 34]. Indeed, the role of the immune system in the pathophysiology of OCD in adults has long been discussed, on the basis of a large amount of data that, in any case, an association between OCD and autoimmunity is conceivable, as shown by a more frequent presence of different autoimmune disorders (systemic lupus erythematosus, or some thyroid diseases) in patients with OCD, as compared with patients with other mental disorders [15, 35–39]. Furthermore, some immune cell alterations were found in adult OCD patients [3, 40].

### 13.3.1 Peripheral Biomarkers of Inflammation in OCD

Recently, growing interest has been given to the detection of inflammatory markers of OCD in easily accessible bodily fluids such as blood and cerebrospinal fluid (CSF). Furthermore, both the innate and the acquired immune systems appear to be involved.

As far as the innate immune system is concerned, much attention has been focused on monocytes, mostly implicated in the first immune response upon infections. Circulating monocytes can be recruited in the central nervous system (CNS) in response to psychosocial stress or brain injury, where they can display their phagocytic activity and release immune mediators such as cytokines [41]. Further peripherally produced cytokines can also enter the CNS boosting the neuroinflammatory response and influencing neurotransmitter availability [42, 43]. Their activity is mainly expressed in the basal ganglia and in the dorsal anterior cingulate cortex, structures that have been related to OCD pathophysiology [44]. The peripheral activation of monocytes may also suggest an activation of microglia, representing the main cells of innate immunity in the CNS. Furthermore, apparently, monocytes and microglia may act similarly in response to external stimuli [45]. In a study of a few years ago [46], the percentage of different subpopulations of monocytes and of their overall number was assessed in blood samples from 102 patients with early-onset OCD, compared to 47 healthy controls (HCs). A flow cytometric analysis was performed to detect different CD profiles on monocyte surface and three different groups of cells were identified: classical, intermediate, and non-classical monocytes [46]. Intermediate monocytes are also referred to as “proinflammatory,” based on their higher production of inflammatory cytokines [47, 48]. In addition, the production of proinflammatory cytokines was evaluated in isolated monocyte cultures both in basal conditions and after exposure to lipopolysaccharide (LPS) or dexamethasone. Interestingly, OCD patients had a significantly higher percentage of total, intermediate, and nonclassical monocytes, compared to controls. Moreover, monocytes of OCD patients released higher amounts of proinflammatory cytokines compared to HCs after exposure to LPS. In

parallel, no significant differences between groups in basal cytokines production and after exposure to dexamethasone were reported [46].

In an effort to evaluate immune system abnormalities in OCD, different subsets of T cells in blood samples have been assessed in unmedicated and comorbidity-free OCD patients and in HCs, applying flow cytometric analysis. A significantly reduced amount of T regulatory (Treg) lymphocytes was observed in OCD subjects, compared to unaffected individuals. Similar results had been previously found in an investigation involving children suffering from OCD-related syndromes [49]. Notably, Treg cells suppress the potentially excessive activity of T helper (Th) lymphocytes, thus playing a critical role in the regulation and homeostasis of immune and autoimmune mechanisms. In particular, these lymphocytes fulfill their function either by inhibiting the production of proinflammatory cytokines or by secreting anti-inflammatory molecules [50, 51]. Those preliminary results further support the immuno-inflammatory theory of OCD and might perhaps lead to new paradigm of peripheral biomarkers detection, bringing substantial advantages in this research field.

Cytokines represent a wide family of glycoproteins produced by several cellular types in different organs and they are involved in mechanisms associated with inflammation, response to infections and autoimmunity. They include interleukins (ILs), chemokines, interferons (IFNs), lymphokines, and tumor necrosis factors (TNFs). Among the most studied cytokines stand IL-6, IL-1β, and TNF-α. Interleukin-6, a pleiotropic factor produced by macrophages and T-cells [52], is involved in several processes such as immune response and hematopoiesis [53], and its overproduction has been associated with several inflammatory diseases [54, 55]. Interestingly, IL-6 was recently found to be increased in drug-naive OCD patients compared to HCs [56], thus posing the question whether it might contribute to OCD pathophysiology. Such finding was consistent with a previous research that highlighted the increased plasma levels not only of IL-6 levels, but also of IL-2, IL-4, IL-10, and TNF-α [57]. Interestingly, antiobsessional drugs may lead to a significant decrease of plasma IL-6 levels [58]. Interleukin-1β is a crucial member of the IL-1 family that has often been the focus of scientific interest due to its role in several autoinflammatory diseases [59]. Along with Il-1α, it acts as a proinflammatory cytokine with pleiotropic effects, including homeostatic processes such as sleep and temperature regulation [60], and it can be released by different cell types, such as neurons, fibroblasts, and several immune cells, like macrophages, mast cells, and microglia [61–64]. In an attempt to shed light on the connection between OCD, immune system, and cognitive dysfunctions, a significant increase in blood IL-1β levels, along with those of IL-6 and TNF-α, was demonstrated in OCD patients [65]. Interestingly, IL-1β was positively related to the Trail Making Test A score, a neuropsychological assessment tool used to evaluate different cognitive skills, thereby suggesting both the role of this cytokine in the pathophysiology of OCD and in cognitive functions of these patients [65]. Again, the available evidence is far from being overall coherent, as a previous work had instead highlighted a decrease in IL-1β plasma levels in OCD [3], while two further studies found no difference [66, 67]. On the other hand, the data around other immune mediators, such as

TNF-α, are more promising. First named by O'Malley et al. in 1962 [68], the actions exerted by this molecule have been progressively unveiled over the years, as it seems to be involved both in the immune response and in cell proliferation and differentiation [69]. It is primarily produced by macrophages and circulating monocytes, and together with its receptors, it is involved in autoimmune and inflammatory processes [27, 58, 70, 71]. Besides the findings reported above, plasma TNF-α levels were found to be increased in drug-naive OCD patients and negatively correlated with the age of onset [58]. Similarly, increased TNF-α levels were demonstrated in children affected by OCD, along with decreased serum IL-12 levels [72]. These findings were suggested to be associated with the role of Th-mediated immune response to psychosocial stress in OCD within this age range [72]. However, a subsequent meta-analysis did not lead to similar findings, while pointing to elevated TNF-α levels in the case of comorbid depression [73]. Interestingly, some OCD patients displayed abnormal changes in the hippocampus, where TNF-α and IL-6 have been found [74, 75]. Soluble TNF receptor-1 and receptor-2 (sTNFR1 and sTNFR2) were found to be increased in their density, thus suggesting an inflammatory condition of mild entity [76]. TNF-α gene and its polymorphisms have also been a subject of investigation. This gene is located on chromosome 6p21.3 that has been suggested to be associated with OCD [73]. In particular, the 308G/A polymorphism may affect TNF-α transcription, albeit data are still inconclusive, and it has been associated with different autoimmune diseases that may also include psychiatric symptoms in their clinical presentation, such as SLE [77–79]. However, even when taken together, these and other data are still overall scant.

Further mediators involved in the mechanisms of autoimmunity may also be involved. A decrease in peripheral T cells was demonstrated in adult subjects with OCD, with a possible association with the severity of symptoms [58, 80]. The levels of different subtypes of lymphocytes may also be altered in OCD, as shown by a decreased activity of natural killer (NK) cells, decreased CD4+ lymphocytes and increased CD8+ lymphocytes [19]. Nevertheless, the immune system should not be considered individually in OCD, as it interfaces and interacts with other equally important systems, such as that mediated by 5-HT [81, 82]. Two crucial immune modulators, that are TNF-α and IFN-γ, may lead to the activation of a key enzyme in the metabolism of tryptophan, the 5-HT precursor, that is indoleamine-2,3-dehydrogenase (IDO) [83]. Indeed, IDO causes a shift in the metabolism of this amino acid, from the production of 5-HT to that of kynurenines [83], thus reducing the synthesis of 5-HT. On the one hand, the latter may cause a condition of vulnerability to different neuropsychiatric disorders including OCD, while on the other hand, kynurenines are tryptophan metabolites that may also exert an excitotoxic effect in the CNS at the level of the glutamate system. Interestingly, glutamate, a ubiquitous excitatory neurotransmitter presents in the brain involved in OCD, has been shown to modulate the function of T lymphocytes [84]. Along this line, a few studies reported increased CSF glutamate levels in OCD patients [85, 86].

### 13.3.2 Microglia Activation in OCD

Preliminary evidence also suggests a possible role of the microglia in OCD.

The main activity of microglia is the protection of CNS homeostasis against triggers of different kinds, as these cells can quickly translate from a resting state to activation, taking part in the mechanisms of both the innate and the adaptive immunity. On the other hand, its excessive alterations may cause several damages to neurons and glias [87]. The inflammation process stimulating the response of the microglia can follow two different pathways of activation, which are M1 and M2. The M1 pathway involves an increase in different cytokines, complement proteins, reactive oxygen species (ROS), and proteinases, while the M2 response enhances tissue remodeling and repair, the expression of angiogenesis factors, and the removal of cellular debris. In any case, microglia activation leads to an increased expression of the translocator protein 18kDA, also known as TPSO, a mitochondrial membrane protein that is considered a key marker of neuroinflammation [29, 88]. Through positron emission tomography (PET), a prominent TSPO activity was detected in the cortico-striatal-thalamo-cortical (CSTC) circuit in OCD patients, that seems to constitute the most specific altered pathway in this condition [32]. Another study analyzing the TSPO distribution volume (VT) as an index of TSPO density to address the matter, reported an increased TSPO VT in most brain regions [29]. Furthermore, TSPO VT in the orbitofrontal cortex was positively associated with the Y-BOCS measure of distress associated with preventing compulsive behaviors. All these findings led to hypothesize that the neuroinflammatory processes in OCD go far beyond the basal ganglia [29].

Finally, it should be emphasized that microglial dysregulation has been documented not only in OCD, but also in TS and PANDAS [89].

### 13.3.3 The Role of Genetics and Fetal-Maternal Immune Interactions

Accumulating evidence shows that OCD and other psychiatric syndromes in children may be related to autoimmune conditions (such as serum autoantibody positivity) and/or, as already mentioned, diseases [90].

Notably, autoimmune conditions are characterized by strong familiarity. One study, enrolling people born between 1940 and 2007 to explore the link between OCD and autoimmune diseases and multigenerational familial correlation, showed that subjects affected by OCD had 43% higher risk of developing autoimmune disorders than those without OCD [30]. In addition, a greater risk was found in relatives of OCD patients compared to family members of HCs. Interestingly, the strongest correlation concerned the mothers (18%) and siblings (16%), while a minor correlation was found with the fathers (8%) [30]. Furthermore, it appears that mothers of OCD children are more likely to develop autoimmune disorders to a greater extent than mothers of children affected by neurologic autoimmune disorders [31].

It has been speculated that the transplacental passage of antibodies from mother to child might have an impact on the neurological and psychiatric development of the fetus. Indeed, maternal immunoglobulins G pass the placenta at the beginning of the second trimester, when the blood-brain barrier (BBB) is not yet fully developed [91]. The transfer of maternal-fetal antibodies has been ascertained for several antibodies, e.g., the presence of contactin-associated protein-like 2 (CASPR2) antibodies has been associated with neurodevelopmental disorders in children. These antibodies cause a decrease in glutamatergic synapses which is then compatible with the autogenic hypothesis of SZ and autism [88, 92]. Specifically, in OCD patients elevated serum levels of ABGA, dopamine receptor of type 2 and lysoganglioside were found compared with HCs. In particular, two antibodies that weighed approximately 55 kDA and 86 kDA were detected [92, 93].

The implications regarding pregnant women with OCD are worth focusing on, as it is a fairly common disorder in pregnancy. In one study, the cord blood of fetuses from mothers with OCD appeared to have higher levels of TNF-α than in fetuses of healthy mothers. This condition might interfere with both neuronal development and the general growth of the fetus. It was actually reported that the children of mothers suffering from OCD had a lower birth weight compared to the children of healthy mothers [94]. Additional studies are warranted to confirm and expand these preliminary hypotheses.

### 13.3.4 Autoimmunity and OCD

OCD-related clinical pictures and dysfunction of the basal ganglia are common findings throughout a spectrum of neuropsychiatric syndromes with a well-demonstrated (e.g., Sydenham chorea) or suspected (pediatric acute-onset neuropsychiatric syndrome, or PANS, and TS) autoimmune etiology [95–98]. Hence, it might be speculated that shared pathophysiological mechanisms underlie those disorders, all manifesting with OCD-like phenotypes. In PANS, robust evidence supports the hypothesis of an induction of ABGA subsequent to molecular mimicry mechanisms involving GABHS [99–101]. As a matter of fact, ABGA exhibit high avidity for certain antigens expressing variable degrees of proteomic similarity with GABHS surface molecules (such as lysoganglioside, tubulin, dopamine receptors of type 1 and 2) and for neuronal glycolytic enzymes (including aldolase C, neuron-specific gamma-enolase, no-neuronal alpha-enolase, and pyruvate kinase M1) [97–99, 102, 103]. Increased odds of ABGA positivity in the serum of patients suffering from these disorders have been widely shown [96, 98, 104, 105]. Nevertheless, the exact causative relationship between ABGA and primary OCD remains elusive. Reports from a large systematic review and meta-analysis reveal that significantly greater proportions of people suffering from OCD are ABGA seropositive compared to different control groups, even after stratifying the analysis by various specifiers (such as age, gender, disease severity, study type). However, no significant differences in ABGA peripheral profile were detected when comparing the primary OCD group with TS, attention-deficit/hyperactivity disorder (ADHD), or PANS

groups. Interestingly, a study examining CSF samples reported a significantly greater proportion of ABGA CSF-positivity in primary OCD patients compared to HCs [28]. Recently, an additional meta-analysis confirmed these observations [92]. Although encouraging, these results are not sufficient to clarify the role of ABGA as putative OCD biomarkers or as mere epiphenomena of the concomitant autoimmune process. In parallel, increasing efforts have been made in trying to better define the functions of immune cells directly involved in autoimmune processes (such as Th cells) in the pathogenesis of OCD. In particular, Th1 and Th17 lymphocytes play a critical part in autoimmune disorders, by producing IL-2, IFN-γ, TNF-α, and IL-17 [106–109]. A study examined the levels of those cytokines in the blood of children with a diagnosis of OCD and in a group of age- and gender-matched controls. Interestingly, significantly higher levels of IL-17, TNF-α, and IL-2 have been found in OCD patients compared to HCs, suggesting a possible implication of Th1 and Th17 lymphocytes in the occurring autoimmune process. Moreover, a lack of correlation between severity and duration of OCS and blood cytokine levels was reported [110]. These results may strengthen the link between autoimmune disorders and OCD, but additional investigations are warranted to expand this evidence.

## 13.4 PANDAS

In 1989 Swedo et al. observed a high prevalence of OCS in children and adolescents with Sydenham chorea and proposed the existence of a link between OCD, basal ganglia, and immunity [111]. Almost 10 years later, the same authors described the clinical characteristics of a new subgroup of 50 patients presenting OCD and tic disorders and fulfilling five diagnostic criteria: (1) presence of OCD and/or a tic disorder; (2) prepubertal onset; (3) episodic course of symptom severity, that is to say an acute symptom onset and relapsing-remitting course; (4) association with GABHS infections; and (5) association with neurological abnormalities [95]. More specifically, the symptomatology also included emotional lability, separation anxiety, night-time fears and bed-time rituals, cognitive deficits, oppositional behaviors, and motoric hyperactivity. As already mentioned, this novel syndrome was named PANDAS [95]. While early-onset and late-onset OCD are characterized by a mean age at onset of, respectively, 11 and 23 years [112], the age at onset for PANDAS is 6–7 years [113]. The temporal association with *Streptococcus pyogenes* infection led to the hypothesis of an autoimmune pathogenetic mechanism similar to that characterizing Sydenham chorea, that represents the neurological manifestation of rheumatic fever, in which streptococcal antibodies cross-react against brain antigens due to a molecular mimicry process [22]. In 2012 PANDAS criteria were modified to describe an expanded clinical entity, PANS, characterized by abrupt, dramatic onset of OCD, or severely restricted food intake, by the concurrent presence of additional neuropsychiatric symptoms with similarly severe and acute onset, and by the exclusion of a known neurologic or medical disorder [114], while entailing that several agents other than Streptococcus might be involved [115]. Therefore,

PANDAS might be considered as a subgroup located within the broader PANS spectrum [22], the latter defining neuropsychiatric conditions triggered by infective, environmental, and metabolic factors [114].

The existence of PANDAS as a distinct entity has been discussed, and its recognition has not met a general agreement [115]. Nonetheless, since its defition, PANDAS has been representing the paradigm of an autoimmune model for OCD, at least in childhood, while encouraging the evaluation of inflammatory, infective, immunologic, and metabolic alterations in patients with acute onset of OCS, neurocognitive and motor symptoms, as well as the evaluation of antibiotics and non-steroidal anti-inflammatory drugs (NSAIDs) as therapeutic strategies [115, 116].

Among the novel theoretic models emerging in psychiatric research and focusing on the immune system, the gut microbiota seems interesting [117]. The gut-brain axis, that is to say, the bidirectional connection occurring between the gut microbiota and the brain, implies a reciprocal influence according to which the diversity in microbiota composition affects brain development and behaviors, and vice versa [118]. A recent study demonstrated the presence of an altered microbiota in PANDAS/PANS patients in comparison to controls, suggesting that GABHS might alter gut microbiota leading to a pro-inflammatory state through the selection of bacterial strains which are associated with gut inflammation and the activation of the immune response. Therefore, according to the gut-brain axis model, an altered bacterial community in the gut would influence behavior, as observed in PANDAS/PANS patients. Some authors also suggested the possibility of studying bacterial biomarkers in these patients, as well as searching for new therapies [22]. On the other hand, since the composition of intestinal flora might be altered even by antibiotics, it has also been suggested that PANDAS might indeed be caused by the antibiotics used to treat the infection, rather than by GABHS [119].

## 13.5 Tourette Syndrome

Originally described in 1885 by the French neurologist Georges Gilles de la Tourette, when he was just a student of Charcot at the Salpétrière hospital in Paris, TS is a neurodevelopmental disorder with a typical onset in childhood and predominantly characterized by motor and vocal (or phonic) tics. Tics may be described as brief, reiterative and involuntary movements or sounds, classically preceded by a sense of urge/discomfort that is relieved after the end of the tic. Their clinical presentation is heterogeneous, and fluctuations in their frequency and severity are typically documented in clinical practice [120]. The following diagnostic criteria must be met to make a diagnosis of TS: (1) the presence of at least two motor tics and one phonic tic, (2) onset of symptoms before 18 years and their persistence for more than 12 months, and (3) these symptoms should not represent a consequence of other neurological disorders, such as encephalitis, stroke and/or other intracranial lesions [7].

Epidemiological data reported an estimated prevalence of TS of 1% [121], with an apparent role of gender, as TS affects about four times more boys than girls [122]. Furthermore, it often presents along with different neuropsychiatric comorbidities, including most frequently OCD and ADHD, but also sleep disturbances and depression [123, 124]

The pathophysiology of TS is far from being completely clarified. The main hypothesis is centered on a multifactorial model encompassing the individual vulnerability (perhaps but not only on a genetic basis) and immunologic/environmental factors [125, 126]. In the genetic field, the focus has primarily been directed towards the *SLITRK1* gene, known for its influence on dendritic growth, and with variants have been associated with TS [127]. Several studies highlighted a dysfunction of the basal ganglia and of the associated CSTC circuitry [128, 129]. Abnormalities of the dopaminergic [130, 131] and serotonergic systems [132] have also been reported.

Nevertheless, the interest of scientists has long been directed to the impact of different infectious agents, mainly but not exclusively GABHS, a common cause of acute pharyngitis in childhood [133], based on the findings of high antistreptolysin O levels and cultural GABHS-positivity in the throat of TS patients [134]. Moreover, the risk for TS was found to be increased in subjects suffering from plurime GABHS infections [135]. GABHS may also be crucial in TS, as patients presenting swings of tics/OCD symptomatology also displayed chronically elevated streptococcal levels than patients having a steady or remitting course of disease [136].

Other infectious agents, such as *M. pneumoniae* [137, 138], *B. burgdorferi* [139], and *C. trachomatis* [140], might also be involved.

GABHS and the other infectious noxae would lead to the activation of an inflammatory response, as documented by higher serum levels of TNF-α and IL-12 [76]. Indeed, either infections or autoimmune response in TS might be caused by a dysregulation of the innate immunity as a predisposing factor, based on different findings, including an increase in monocyte levels [141]. The existence of an immune/inflammatory response in TS is also critical for its therapeutic implications. Anecdotally, a patient affected by chronic TS was treated, as an add-on to the antibiotic prophylaxis, with celecoxib [142], a NSAID belonging to the group of ciclooxigenase-2 (COX-2) selective inhibitors [143]. This treatment was successful in improving both the motor symptoms and the altered behavior (specifically social retreat and aggressivity). More interestingly, discontinuation of the drug led to a heavy deterioration of tics, and its re-administration caused relevant benefits [142]. That and similar evidence would suggest new frontiers of treatment strategies, going beyond the currently most used treatments, including comprehensive behavioral interventions for tics and different antipsychotics, such as haloperidol, aripiprazole, and risperidone [144, 145].

### 13.5.1 Genetic Vulnerability to an Aberrant Autoimmune Response in TS

As already mentioned, it is likely that a susceptibility due to a genetic basis may represent a key factor in the genesis of TS. Most important, an increasing amount of evidence supports the hypothesis that the expression of specific genes may alter the mechanisms of autoimmunity in these patients.

A role of the expression of genes linked to catecholamines in TS has been suggested, as it was documented an overexpression dopamine receptors of type 22, of histamine receptors of type 3, of brain-derived neurotrophic factor (BDNF), and others [131, 146, 147]. Furthermore, since dopamine seems to exert a regulatory function on immunity cells, such as different subtypes of T lymphocytes [148], it is possible that, either directly or indirectly, it might contribute to the onset of TS, a although more and substantial evidence on the matter is warranted. Interestingly, the genetic expression of catecholamines has been associated with the severity of TS symptomatology [147].

Similarly, a relationship between the severity of the clinical picture and the genes associated with the cholinergic system and gamma-aminobutyric acid (GABA) has been reported [149]. It is noteworthy that acetylcholine and its receptors play a role in the modulation of both B and T lymphocytes [150]. Furthermore, these receptors also regulate the release of dopamine, due to their expression in dopaminergic and GABAergic neurons of the striatum [150], thus suggesting that their dysregulation may take a part yet to be unveiled in TS. Regarding the role of GABA, the main findings so far suggest a negative correlation between the GABA receptor-associated protein (GABARAP) and the severity of tics [149]. GABA and its receptors, also present in macrophages and lymphocytes, display several effects on the modulation of different immunity cells, such as neutrophils [151].

### 13.5.2 Microglia Activation in TS

An aberrant activation of microglia in the striatum of TS patients represents one of the main pieces of evidence collected so far. Specifically, a postmortem study demonstrated an increased expression of CD45+ in microglia cells and, as such, an hyperactivation of these cells in the striatum [152]. Consistently with this finding, a PET study using (11)C-[R]-PK11195 that binds to translocator protein by microglia cells following their activation demonstrated an increased binding of this ligand in the caudate nuclei of a pool of children affected by TS and PANDAS [153]. However, the PANDAS group displayed an increased neuroinflammation induced by microglia activation in both bilateral lentiform nucleus and bilateral caudate, while the TS group displayed an inflammatory activation of bilateral caudate nuclei alone. Such findings were not detected in the adults. These data led the authors to suggest the involvement of different inflammatory pathways in the two disorders [153]. In addition, data gathered from mice models appear to strengthen

the relationship between microglia and the pathogenesis of TS and further suggest hyperreactivity to noxae in these patients [89, 154].

### 13.5.3 Effector Molecules of Immunity/Inflammation and Their Role in TS

Chemokines, cytokines, and adhesion molecules are a heterogenous class of molecules whose role in the activation and modulation of the innate and the adaptive immune systems is long known. In particular, cytokines and chemokines are heavily involved in a wide range of functions, such as cell growth, differentiation, trafficking, and regulation following insults of different etiology, and also determine the type of response activated (humoral, cell-mediated, cytotoxic, allergic) [155]. Current findings also point to a proinflammatory state in TS, where the role of such mediators appears to be crucial. Interestingly, plasma TNF-α and IL-12 levels, along with those of other cytokines (IL-6, IL-17) further increased during the exacerbation of the symptoms in a sample of children affected by TS but not OCD [76, 156]. Interestingly, it is worth noting that drug-naive patients also displayed higher TNF-α levels than controls [156].

The comorbidity of TS and OCD might involve different immunological pathways than patients affected by TS alone. Indeed, only in the former IL-2 and IL-12 plasma levels were higher in comparison to the control group, while no significant differences were detected in the other cytokines examined (IL-1β, IL-2, and IL-6) [157]. Noteworthy, TS and OCD seem to show opposite patterns in terms of cytokine secretion, given the above findings and the evidence of decreased TNF-α and IL-1β and increased IL-6 levels [66, 67, 158], thus suggesting how the immunological pathways may differ in these disorders. Finally, a positive correlation was also found with plasma levels of neopterin. Neopterin, a metabolite of guanosine triphosphate, mainly known as a biomarker of cell-mediated immunity [159], resulted significantly higher in two different studies involving adolescents and/or children affected by TS [141, 160].

### 13.5.4 Regulation/Dysregulation of Immunity Cells in TS: Which Ones and How?

Studies targeting immunity cell subpopulations analysis in TS led to a mix of intriguing results. First of all, an alteration in these cells' number or function may be also strictly related to the clinical picture, as in the case of Treg cells, that have been proposed as important mediators in the pathogenesis of TS and chronic tic disorder [161]. This subpopulation of T-cells is fundamental in the mechanisms of autoimmunity, inflammation, and allergic response [162]. A correlation between their decreased number and severity of TS symptoms has been documented in one study [163]. Moreover, the association between dopamine and T cells might also be closer than originally hypothesized, as patients affected by TS showed not only a

higher mRNA expression of dopamine receptors 1-5 in peripheral blood lymphocytes, but also a significant correlation with the severity of compulsive symptoms [164]. The investigation of several lymphocyte surface markers in a sample of 20 adults affected by TS reported a significant difference in CD69+/CD22+ B cells and in CD95+/CD4+ T cells, thus leading to hypothesize an increase in peripheral immune activity in this disorder [165]. As a matter of fact, CD69 represents a marker of early activation of lymphocytes [166], while CD95 is strictly associated with T-cells death [167]. Therefore, their increase, as in the case of the previous research, suggests an augmented removal of activated cells in the periphery and, so, an increase in peripheral immune activity.

However, literature is still controversial, and in a recent study, no significant phenotypic differences on higher levels of inflammatory markers in the CSF of a sample of children affected by TS and positive to Streptococci were demonstrated [168].

## 13.6 Future Directions

Data collected up to now, although requiring more solid and widespread evidence, seem to suggest a key role of immune alterations in the pathophysiology of OCD, with complex mechanisms that are yet to be fully explained. Available findings would suggest that OCD and related conditions are deeply linked to immune system alterations, often triggered by several agents, as stress or infectious insults. Nonetheless, from a therapeutic perspective, a deeper understanding of the complex interplay between nervous and immune systems, as well as of immune alterations detectable in patients, might help to develop new therapeutic strategies, taking into account inflammatory and immunologic mechanisms.

At the moment, immunotherapy, antibiotics prophylaxis and administration of oral penicillin, plasmapheresis, and intravenous immunoglobulins have all been proposed as therapeutic options in reducing the symptoms of OCD and related conditions, but results remain controversial to date, especially in adult patients [14, 169].

In comparison to placebo, cefdinir, a β-lactam antibiotic, led to improvements of both tic and OCD symptoms, although the effects were non-statistically significant. Nonetheless, the authors underlined how β-lactam antibiotics might have neuroprotective properties, beyond antimicrobial ones [170]. As far as augmentation strategies are concerned, a clinical trial on the efficacy of celecoxib as an adjunct in the treatment of OCD showed that its combination with fluoxetine led to a more robust decrease of OCS than fluoxetine plus placebo [80]. Similar results were observed with a fluvoxamine-celecoxib combination [171].

## 13.7 Conclusions

The spectrum of OCD disorders including among the others OCD, PANDAS, and TS might recognize common biological underpinnings also involving an altered immune response due to noxae of different kinds. This immune dysregulation is likely to represent a part of a complex etiopathogenetic mosaic. According to available data, the molecules and mediators of the immune response in these neuropsychiatric disorders might be similar, but still differ\according to patients' age, genetics or environmental insults. Their specific role, the brain areas that are mostly involved and how they might correlate with the severity of the clinical picture is another topic of interest, mainly but not only in the case of PANDAS that was recently recognized, as compared with OCD and TS, and is characterized by a wide range of symptoms.

Animal and human studies (both in adults and children patients) seem to support such a statement, albeit nowadays the evidence is still inconclusive, given the paucity of the size of the samples and of the amount of research. The hope is that it will be possible to clarify which clinical features, symptom clusters or dimensions might be related to specific immunologic/inflammatory alterations. Therefore, further effort is desirable in the near future to unveil the peculiar pathogenetic mechanisms behind these neuropsychiatric disorders. The potential scenario of therapeutic options that may consequently unfold is intriguing, as drugs used today for other purposes, as well as new compounds, might appear in the horizon as valid tools.

## References

1. Marazziti D, Mucci F, Lombardi A, Falaschi V, Dell'Osso L. The cytokine profile of OCD: pathophysiological insights. Int J Interferon Cytokine Mediat Res. 2015;7:35–42. https://doi.org/10.2147/ijicmr.s76710.
2. Kerr D, Krishnan C, Pucak ML, Carmen J. The immune system and neuropsychiatric diseases. Int Rev Psychiatry. 2005;17(6):443–9. https://doi.org/10.1080/0264830500381435.
3. Brambilla F, Bellodi L, Perna G. Plasma levels of tumor necrosis factor-alpha in patients with panic disorder: effect of alprazolam therapy. Psychiatry Res. 1999;89(1):21–7. https://doi.org/10.1016/s0165-1781(99)00091-8.
4. Rapaport MH, Stein MB. Serum interleukin-2 and soluble interleukin-2 receptor levels in generalized social phobia. Anxiety. 1994;1(2):50–3. https://doi.org/10.1002/anxi.3070010203.
5. Dell'Osso L, Carmassi C, Mucci F, Marazziti D. Depression, serotonin and tryptophan. Curr Pharm Des. 2016;22(8):949–54. https://doi.org/10.2174/1381612822666151214104826.
6. Fawcett EJ, Power H, Fawcett JM. Women are at greater risk of OCD than men: a meta-analytic review of OCD prevalence worldwide. J Clin Psychiatry. 2020;81(4):19r13085. https://doi.org/10.4088/JCP.19r13085.
7. American Psychiatric Association. Diagnostic and statistical manual of mental disorders. 5th ed. Arlington, VA: American Psychiatric Association; 2013.
8. Pallanti S, Hollander E, Bienstock C, Koran L, Leckman J, Marazziti D, et al. Treatment non-response in OCD: methodological issues and operational definitions. Int J Neuropsychopharmacol. 2002;5(2):181–91. https://doi.org/10.1017/S1461145702002900.

9. Marazziti D, Picchetti M, Baroni S, Ceresoli D, Consoli G, Catena Dell'Osso M. Current pharmacological and non pharmacological treatments for obsessive-compulsive disorder. J Psychopathol. 2012;18(1):5–18.
10. Denys D, Zohar J, Westenberg HG. The role of dopamine in obsessive-compulsive disorder: preclinical and clinical evidence. J Clin Psychiatry. 2004;65(Suppl 14):11–7.
11. Pittenger C, Bloch MH, Williams K. Glutamate abnormalities in obsessive compulsive disorder: neurobiology, pathophysiology, and treatment. Pharmacol Ther. 2011;132(3):314–32. https://doi.org/10.1016/j.pharmthera.2011.09.006.
12. Marazziti D, Albert U, Mucci F, Piccinni A. The glutamate and the immune systems: new targets for the pharmacological treatment of OCD. Curr Med Chem. 2018;25(41):5731–8. https://doi.org/10.2174/0929867324666171108152035.
13. Murphy ML, Pichichero ME. Prospective identification and treatment of children with pediatric autoimmune neuropsychiatric disorder associated with group A streptococcal infection (PANDAS). Arch Pediatr Adolesc Med. 2002;156(4):356–61. https://doi.org/10.1001/archpedi.156.4.356.
14. Murphy TK, Sajid MW, Goodman WK. Immunology of obsessive-compulsive disorder. Psychiatr Clin North Am. 2006;29(2):445–69. https://doi.org/10.1016/j.psc.2006.02.003.
15. da Rocha FF, Correa H, Teixeira AL. Obsessive-compulsive disorder and immunology: a review. Prog Neuropsychopharmacol Biol Psychiatry. 2008;32(5):1139–46. https://doi.org/10.1016/j.pnpbp.2007.12.026.
16. Teixeira AL, Rodrigues DH, Marques AH, Miguel EC, Fontenelle LF. Searching for the immune basis of obsessive-compulsive disorder. Neuroimmunomodulation. 2014;21(2-3):152–8. https://doi.org/10.1159/000356554.
17. Marazziti D, Mucci F, Fontenelle LF. Immune system and obsessive-compulsive disorder. Psychoneuroendocrinology. 2018;93:39–44. https://doi.org/10.1016/j.psyneuen.2018.04.013.
18. Cosco TD, Pillinger T, Emam H, Solmi M, Budhdeo S, Matthew Prina A, et al. Immune aberrations in obsessive-compulsive disorder: a systematic review and meta-analysis. Mol Neurobiol. 2019;56(7):4751–9. https://doi.org/10.1007/s12035-018-1409-x.
19. Marazziti D, Presta S, Pfanner C, Gemignani A, Rossi A, Sbrana S, et al. Immunological alterations in adult obsessive-compulsive disorder. Biol Psychiatry. 1999;46(6):810–4. https://doi.org/10.1016/s0006-3223(98)00371-0.
20. Lynch NE, Deiratany S, Webb DW, McMenamin JB. PANDAS (Paediatric Autoimmune Neuropsychiatric Disorder Associated with Streptococcal Infection). Ir Med J. 2006;99(5):155.
21. Swedo SE, Seidlitz J, Kovacevic M, Latimer ME, Hommer R, Lougee L, et al. Clinical presentation of pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections in research and community settings. J Child Adolesc Psychopharmacol. 2015;25(1):26–30. https://doi.org/10.1089/cap.2014.0073.
22. Quagliariello A, Del Chierico F, Russo A, Reddel S, Conte G, Lopetuso LR, et al. Gut microbiota profiling and gut-brain crosstalk in children affected by pediatric acute-onset neuropsychiatric syndrome and pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections. Front Microbiol. 2018;9:675. https://doi.org/10.3389/fmicb.2018.00675.
23. Pavone P, Falsaperla R, Cacciaguerra G, Sapuppo A, Chiaramonte R, Lubrano R, et al. PANS/PANDAS: Clinical experience in IVIG treatment and state of the art in rehabilitation approaches. NeuroSci. 2020;1(2):75–84. https://doi.org/10.3390/neurosci1020007.
24. Cavanna AE, Termine C. Tourette syndrome. Adv Exp Med Biol. 2012;724:375–83. https://doi.org/10.1007/978-1-4614-0653-2_28.
25. Martino D, Zis P, Buttiglione M. The role of immune mechanisms in Tourette syndrome. Brain Res. 2015;1617:126–43. https://doi.org/10.1016/j.brainres.2014.04.027.
26. Hsu CJ, Wong LC, Lee WT. Immunological dysfunction in Tourette syndrome and related disorders. Int J Mol Sci. 2021;22(2):853. https://doi.org/10.3390/ijms22020853.

27. Fluitman S, Denys D, Vulink N, Schutters S, Heijnen C, Westenberg H. Lipopolysaccharide-induced cytokine production in obsessive-compulsive disorder and generalized social anxiety disorder. Psychiatry Res. 2010;178(2):313–6. https://doi.org/10.1016/j.psychres.2009.05.008.
28. Pearlman DM, Vora HS, Marquis BG, Najjar S, Dudley LA. Anti-basal ganglia antibodies in primary obsessive-compulsive disorder: systematic review and meta-analysis. Br J Psychiatry. 2014;205(1):8–16. https://doi.org/10.1192/bjp.bp.113.137018.
29. Attwells S, Setiawan E, Wilson AA, Rusjan PM, Mizrahi R, Miler L, et al. Inflammation in the neurocircuitry of obsessive-compulsive disorder. JAMA Psychiatry. 2017;74(8):833–40. https://doi.org/10.1001/jamapsychiatry.2017.1567.
30. Mataix-Cols D, Frans E, Pérez-Vigil A, Kuja-Halkola R, Gromark C, Isomura K, et al. A total-population multigenerational family clustering study of autoimmune diseases in obsessive-compulsive disorder and Tourette's/chronic tic disorders. Mol Psychiatry. 2018;23(7):1652–8. https://doi.org/10.1038/mp.2017.215.
31. Jones HF, Han VX, Patel S, Gloss BS, Soler N, Ho A, et al. Maternal autoimmunity and inflammation are associated with childhood tics and obsessive-compulsive disorder: transcriptomic data show common enriched innate immune pathways. Brain Behav Immun. 2021;94:308–17. https://doi.org/10.1016/j.bbi.2020.12.035.
32. Meyer J. Inflammation, obsessive-compulsive disorder, and related disorders. Curr Top Behav Neurosci. 2021;49:31–53. https://doi.org/10.1007/7854_2020_210.
33. Rao NP, Reddy MS, Reddy JY. Is there a role for immunological mechanisms in etiopathogenesis of obsessive-compulsive disorder? Indian J Psychol Med. 2013;35(1):1–3. https://doi.org/10.4103/0253-7176.112192.
34. Gerentes M, Pelissolo A, Rajagopal K, Tamouza R, Hamdani N. Obsessive-Compulsive Disorder: autoimmunity and neuroinflammation. Curr Psychiatry Rep. 2019;21(8):78. https://doi.org/10.1007/s11920-019-1062-8.
35. Miguel EC, Stein MC, Rauch SL, O'Sullivan RL, Stern TA, Jenike MA. Obsessive-compulsive disorder in patients with multiple sclerosis. J Neuropsychiatry Clin Neurosci. 1995;7(4):507–10. https://doi.org/10.1176/jnp.7.4.507.
36. Placidi GP, Boldrini M, Patronelli A, Fiore E, Chiovato L, Perugi G, et al. Prevalence of psychiatric disorders in thyroid diseased patients. Neuropsychobiology. 1998;38(4):222–5. https://doi.org/10.1159/000026545.
37. Dinn WM, Harris CL, McGonigal KM, Raynard RC. Obsessive-compulsive disorder and immunocompetence. Int J Psychiatry Med. 2001;31(3):311–20. https://doi.org/10.2190/F0BA-BN4F-61KA-UD99.
38. Hoekstra PJ, Minderaa RB. Tic disorders and obsessive-compulsive disorder: is autoimmunity involved? Int Rev Psychiatry. 2005;17(6):497–502. https://doi.org/10.1080/02646830500382003.
39. Tinelli E, Francia A, Quartuccio EM, Morreale M, Contessa GM, Pascucci S, et al. Structural brain MR imaging changes associated with obsessive-compulsive disorder in patients with multiple sclerosis. AJNR Am J Neuroradiol. 2013;34(2):305–9. https://doi.org/10.3174/ajnr.A3210.
40. Marazziti D, Mungai F, Masala I, Baroni S, Vivarelli L, Ambrogi F, et al. Normalisation of immune cell imbalance after pharmacological treatments of patients suffering from obsessive-compulsive disorder. J Psychopharmacol. 2009;23(5):567–73. https://doi.org/10.1177/0269881108089605.
41. Wohleb ES, McKim DB, Sheridan JF, Godbout JP. Monocyte trafficking to the brain with stress and inflammation: a novel axis of immune-to-brain communication that influences mood and behavior. Front Neurosci. 2015;8:447. https://doi.org/10.3389/fnins.2014.00447.
42. Dunn AJ. Effects of cytokines and infections on brain neurochemistry. Clin Neurosci Res. 2006;6(1-2):52–68. https://doi.org/10.1016/j.cnr.2006.04.002.
43. Felger JC, Lotrich FE. Inflammatory cytokines in depression: neurobiological mechanisms and therapeutic implications. Neuroscience. 2013;246:199–229. https://doi.org/10.1016/j.neuroscience.2013.04.060.

44. Pauls DL, Abramovitch A, Rauch SL, Geller DA. Obsessive-compulsive disorder: an integrative genetic and neurobiological perspective. Nat Rev Neurosci. 2014;15(6):410–24. https://doi.org/10.1038/nrn3746.
45. Prinz M, Priller J. Microglia and brain macrophages in the molecular age: from origin to neuropsychiatric disease. Nat Rev Neurosci. 2014;15(5):300–12. https://doi.org/10.1038/nrn3722.
46. Rodríguez N, Morer A, González-Navarro EA, Serra-Pages C, Boloc D, Torres T, et al. Inflammatory dysregulation of monocytes in pediatric patients with obsessive-compulsive disorder. J Neuroinflammation. 2017;14(1):261. https://doi.org/10.1186/s12974-017-1042-z.
47. Wong KL, Yeap WH, Tai JJ, Ong SM, Dang TM, Wong SC. The three human monocyte subsets: implications for health and disease. Immunol Res. 2012;53(1-3):41–57. https://doi.org/10.1007/s12026-012-8297-3.
48. Ziegler-Heitbrock L, Hofer TP. Toward a refined definition of monocyte subsets. Front Immunol. 2013;4:23. https://doi.org/10.3389/fimmu.2013.00023.
49. Subbanna M, Shivakumar V, Jose D, Venkataswamy M, Debnath M, Ravi V, et al. Reduced T cell immunity in unmedicated, comorbidity-free obsessive-compulsive disorder: an immunophenotyping study. J Psychiatr Res. 2021;137:521–4. https://doi.org/10.1016/j.jpsychires.2021.03.035.
50. Sakaguchi S. Naturally arising CD4+ regulatory t cells for immunologic self-tolerance and negative control of immune responses. Annu Rev Immunol. 2004;22:531–62. https://doi.org/10.1146/annurev.immunol.21.120601.141122.
51. Askenasy N, Kaminitz A, Yarkoni S. Mechanisms of T regulatory cell function. Autoimmun Rev. 2008;7(5):370–5. https://doi.org/10.1016/j.autrev.2008.03.001.
52. Sawada M, Imamura K, Nagatsu T (2006) Role of cytokines in inflammatory process in Parkinson's disease. J Neural Transm Suppl (70):373-381. https://doi.org/10.1007/978-3-211-45295-0_57
53. Kishimoto T. IL-6: from its discovery to clinical applications. Int Immunol. 2010;22(5):347–52. https://doi.org/10.1093/intimm/dxq030.
54. Hunter CA, Jones SA. IL-6 as a keystone cytokine in health and disease. Nat Immunol. 2015;16(5):448–57. https://doi.org/10.1038/ni.3153; Erratum in: Nat Immunol. 2017 Oct 18;18(11):1271
55. Tanaka T, Narazaki M, Masuda K, Kishimoto T. Regulation of IL-6 in immunity and diseases. Adv Exp Med Biol. 2016;941:79–88. https://doi.org/10.1007/978-94-024-0921-5_4.
56. Jose D, Dinakaran D, Shivakumar V, Subbanna M, Reddy YCJ, Venkatasubramanian G, et al. Plasma IL-6 levels in unmedicated, comorbidity free obsessive-compulsive disorder. Int J Psychiatry Clin Pract. 2021;25(4):437–40. https://doi.org/10.1080/13651501.2021.1937657.
57. Rao NP, Venkatasubramanian G, Ravi V, Kalmady S, Cherian A, Yc JR. Plasma cytokine abnormalities in drug-naïve, comorbidity-free obsessive-compulsive disorder. Psychiatry Res. 2015;229(3):949–52. https://doi.org/10.1016/j.psychres.2015.07.009.
58. Konuk N, Tekin IO, Ozturk U, Atik L, Atasoy N, Bektas S, et al. Plasma levels of tumor necrosis factor-alpha and interleukin-6 in obsessive compulsive disorder. Mediators Inflamm. 2007;2007:65704. https://doi.org/10.1155/2007/65704.
59. Dinarello CA. Interleukin-1 in the pathogenesis and treatment of inflammatory diseases. Blood. 2011;117(14):3720–32. https://doi.org/10.1182/blood-2010-07-273417.
60. Dinarello CA. Biologic basis for interleukin-1 in disease. Blood. 1996;87(6):2095–147.
61. Watkins LR, Maier SF, Goehler LE. Immune activation: the role of pro-inflammatory cytokines in inflammation, illness responses and pathological pain states. Pain. 1995;63(3):289–302. https://doi.org/10.1016/0304-3959(95)00186-7.
62. Sommer C, Kress M. Recent findings on how proinflammatory cytokines cause pain: peripheral mechanisms in inflammatory and neuropathic hyperalgesia. Neurosci Lett. 2004;361(1-3):184–7. https://doi.org/10.1016/j.neulet.2003.12.007.

63. Clark AK, D'Aquisto F, Gentry C, Marchand F, McMahon SB, Malcangio M. Rapid co-release of interleukin 1beta and caspase 1 in spinal cord inflammation. J Neurochem. 2006;99(3):868–80. https://doi.org/10.1111/j.1471-4159.2006.04126.x.
64. Thacker MA, Clark AK, Marchand F, McMahon SB. Pathophysiology of peripheral neuropathic pain: immune cells and molecules. Anesth Analg. 2007;105(3):838–47. https://doi.org/10.1213/01.ane.0000275190.42912.37.
65. Karagüzel EÖ, Arslan FC, Uysal EK, Demir S, Aykut DS, Tat M, et al. Blood levels of interleukin-1 beta, interleukin-6 and tumor necrosis factor-alpha and cognitive functions in patients with obsessive compulsive disorder. Compr Psychiatry. 2019;89:61–6. https://doi.org/10.1016/j.comppsych.2018.11.013.
66. Monteleone P, Catapano F, Fabrazzo M, Tortorella A, Maj M. Decreased blood levels of tumor necrosis factor-alpha in patients with obsessive-compulsive disorder. Neuropsychobiology. 1998;37(4):182–5. https://doi.org/10.1159/000026500.
67. Denys D, Fluitman S, Kavelaars A, Heijnen C, Westenberg H. Decreased TNF-alpha and NK activity in obsessive-compulsive disorder. Psychoneuroendocrinology. 2004;29(7):945–52. https://doi.org/10.1016/j.psyneuen.2003.08.008.
68. O'Malley WE, Achinstein B, Shear MJ. Journal of the National Cancer Institute, vol. 29, 1962: action of bacterial polysaccharide on tumors. II. Damage of sarcoma 37 by serum of mice treated with Serratia marcescens polysaccharide, and induced tolerance. Nutr Rev. 1988;46(11):389–91. https://doi.org/10.1111/j.1753-4887.1988.tb05376.x.
69. Hayashi K, Piras V, Tabata S, Tomita M, Selvarajoo K. A systems biology approach to suppress TNF-induced proinflammatory gene expressions. Cell Commun Signal. 2013;11:84. https://doi.org/10.1186/1478-811X-11-84.
70. Dudbridge F. Pedigree disequilibrium tests for multilocus haplotypes. Genet Epidemiol. 2003;25(2):115–21. https://doi.org/10.1002/gepi.10252.
71. Haddy N, Sass C, Maumus S, Marie B, Droesch S, Siest G, et al. Biological variations, genetic polymorphisms and familial resemblance of TNF-alpha and IL-6 concentrations: STANISLAS cohort. Eur J Hum Genet. 2005;13(1):109–17. https://doi.org/10.1038/sj.ejhg.5201294.
72. Çolak Sivri R, Bilgiç A, Kılınç İ. Cytokine, chemokine and BDNF levels in medication-free pediatric patients with obsessive-compulsive disorder. Eur Child Adolesc Psychiatry. 2018;27(8):977–84. https://doi.org/10.1007/s00787-017-1099-3.
73. Gray SM, Bloch MH. Systematic review of proinflammatory cytokines in obsessive-compulsive disorder. Curr Psychiatry Rep. 2012;14(3):220–8. https://doi.org/10.1007/s11920-012-0272-0.
74. Cavedini P, Gorini A, Bellodi L. Understanding obsessive-compulsive disorder: focus on decision making. Neuropsychol Rev. 2006;16(1):3–15. https://doi.org/10.1007/s11065-006-9001-y.
75. de Geus F, Denys DA, Sitskoorn MM, Westenberg HG. Attention and cognition in patients with obsessive-compulsive disorder. Psychiatry Clin Neurosci. 2007;61(1):45–53. https://doi.org/10.1111/j.1440-1819.2007.01609.x.
76. Leckman JF, Katsovich L, Kawikova I, Lin H, Zhang H, Krönig H, et al. Increased serum levels of interleukin-12 and tumor necrosis factor-alpha in Tourette's syndrome. Biol Psychiatry. 2005;57(6):667–73. https://doi.org/10.1016/j.biopsych.2004.12.004.
77. Zúñiga J, Vargas-Alarcón G, Hernández-Pacheco G, Portal-Celhay C, Yamamoto-Furusho JK, Granados J. Tumor necrosis factor-alpha promoter polymorphisms in Mexican patients with systemic lupus erythematosus (SLE). Genes Immun. 2001;2(7):363–6. https://doi.org/10.1038/sj.gene.6363793.
78. Fontenelle LF, Barbosa IG, Luna JV, de Sousa LP, Abreu MN, Teixeira AL. A cytokine study of adult patients with obsessive-compulsive disorder. Compr Psychiatry. 2012;53(6):797–804. https://doi.org/10.1016/j.comppsych.2011.12.007.

79. Scalzo P, Kümmer A, Cardoso F, Teixeira AL. Increased serum levels of soluble tumor necrosis factor-alpha receptor-1 in patients with Parkinson's disease. J Neuroimmunol. 2009;216(1-2):122–5. https://doi.org/10.1016/j.jneuroim.2009.08.001.
80. Sayyah M, Boostani H, Pakseresht S, Malayeri A. A preliminary randomized double-blind clinical trial on the efficacy of celecoxib as an adjunct in the treatment of obsessive-compulsive disorder. Psychiatry Res. 2011;189(3):403–6. https://doi.org/10.1016/j.psychres.2011.01.019.
81. Marazziti D, Consoli G, Baroni S, Catena Dell'Osso M. Past, present and future drugs for the treatment of obsessive-compulsive disorder. Curr Med Chem. 2010;17(29):3410–21. https://doi.org/10.2174/092986710793176384.
82. Marazziti D, Consoli G, Masala I, Catena Dell'Osso M, Baroni S. Latest advancements on serotonin and dopamine transporters in lymphocytes. Mini Rev Med Chem. 2010;10(1):32–40. https://doi.org/10.2174/138955710791112587.
83. Lestage J, Verrier D, Palin K, Dantzer R. The enzyme indoleamine 2,3-dioxygenase is induced in the mouse brain in response to peripheral administration of lipopolysaccharide and superantigen. Brain Behav Immun. 2002;16(5):596–601. https://doi.org/10.1016/s0889-1591(02)00014-4.
84. Pacheco R, Gallart T, Lluis C, Franco R. Role of glutamate on T-cell mediated immunity. J Neuroimmunol. 2007;185(1-2):9–19. https://doi.org/10.1016/j.jneuroim.2007.01.003.
85. Chakrabarty K, Bhattacharyya S, Christopher R, Khanna S. Glutamatergic dysfunction in OCD. Neuropsychopharmacology. 2005;30(9):1735–40. https://doi.org/10.1038/sj.npp.1300733.
86. Bhattacharyya S, Khanna S, Chakrabarty K, Mahadevan A, Christopher R, Shankar SK. Anti-brain autoantibodies and altered excitatory neurotransmitters in obsessive-compulsive disorder. Neuropsychopharmacology. 2009;34(12):2489–96. https://doi.org/10.1038/npp.2009.77.
87. van Rossum D, Hanisch UK. Microglia. Metab Brain Dis. 2004;19(3-4):393–411. https://doi.org/10.1023/b:mebr.0000043984.73063.d8.
88. Werry EL, Bright FM, Piguet O, Ittner LM, Halliday GM, Hodges JR, et al. Recent developments in TSPO PET imaging as a biomarker of neuroinflammation in neurodegenerative disorders. Int J Mol Sci. 2019;20(13):3161. https://doi.org/10.3390/ijms20133161.
89. Frick L, Pittenger C. Microglial dysregulation in OCD, Tourette syndrome, and PANDAS. J Immunol Res. 2016;2016:8606057. https://doi.org/10.1155/2016/8606057.
90. Wang LY, Chiang JH, Chen SF, Shen YC. Systemic autoimmune diseases are associated with an increased risk of bipolar disorder: a nationwide population-based cohort study. J Affect Disord. 2018;227:31–7. https://doi.org/10.1016/j.jad.2017.10.027.
91. Palmeira P, Quinello C, Silveira-Lessa AL, Zago CA, Carneiro-Sampaio M. IgG placental transfer in healthy and pathological pregnancies. Clin Dev Immunol. 2012;2012:985646. https://doi.org/10.1155/2012/985646.
92. Hansen N, Luedecke D, Malchow B, Lipp M, Vogelgsang J, Timäus C, et al. Autoantibody-associated psychiatric syndromes in children: link to adult psychiatry. J Neural Transm (Vienna). 2021;128(6):735–47. https://doi.org/10.1007/s00702-021-02354-8.
93. Morer A, Lázaro L, Sabater L, Massana J, Castro J, Graus F. Antineuronal antibodies in a group of children with obsessive-compulsive disorder and Tourette syndrome. J Psychiatr Res. 2008;42(1):64–8. https://doi.org/10.1016/j.jpsychires.2006.09.010.
94. Uguz F, Onder Sonmez E, Sahingoz M, Gokmen Z, Basaran M, Gezginc K, et al. Neuroinflammation in the fetus exposed to maternal obsessive-compulsive disorder during pregnancy: a comparative study on cord blood tumor necrosis factor-alpha levels. Compr Psychiatry. 2014;55(4):861–5. https://doi.org/10.1016/j.comppsych.2013.12.018.
95. Swedo SE, Leonard HL, Garvey M, Mittleman B, Allen AJ, Perlmutter S, et al. Pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections: clinical description of the first 50 cases. Am J Psychiatry. 1998;155(2):264–71. https://doi.org/10.1176/ajp.155.2.264; Erratum in: Am J Psychiatry. 1998 Apr;155(4):578

96. Swedo SE, Leonard HL, Rapoport JL. The pediatric autoimmune neuropsychiatric disorders associated with streptococcal infection (PANDAS) subgroup: separating fact from fiction. Pediatrics. 2004;113(4):907–11. https://doi.org/10.1542/peds.113.4.907.
97. Kirvan CA, Cox CJ, Swedo SE, Cunningham MW. Tubulin is a neuronal target of autoantibodies in Sydenham's chorea. J Immunol. 2007;178(11):7412–21. https://doi.org/10.4049/jimmunol.178.11.7412.
98. Brimberg L, Benhar I, Mascaro-Blanco A, Alvarez K, Lotan D, Winter C, et al. Behavioral, pharmacological, and immunological abnormalities after streptococcal exposure: a novel rat model of Sydenham chorea and related neuropsychiatric disorders. Neuropsychopharmacology. 2012;37(9):2076–87. https://doi.org/10.1038/npp.2012.56.
99. Kirvan CA, Swedo SE, Snider LA, Cunningham MW. Antibody-mediated neuronal cell signaling in behavior and movement disorders. J Neuroimmunol. 2006;179(1-2):173–9. https://doi.org/10.1016/j.jneuroim.2006.06.017.
100. Martino D, Church A, Giovannoni G. Are antibasal ganglia antibodies important, and clinically useful? Pract Neurol. 2007;7(1):32–41.
101. Dale RC, Brilot F. Autoimmune basal ganglia disorders. J Child Neurol. 2012;27(11):1470–81. https://doi.org/10.1177/0883073812451327.
102. Kirvan CA, Swedo SE, Heuser JS, Cunningham MW. Mimicry and autoantibody-mediated neuronal cell signaling in Sydenham chorea. Nat Med. 2003;9(7):914–20. https://doi.org/10.1038/nm892.
103. Dale RC, Candler PM, Church AJ, Wait R, Pocock JM, Giovannoni G. Neuronal surface glycolytic enzymes are autoantigen targets in post-streptococcal autoimmune CNS disease. J Neuroimmunol. 2006;172(1-2):187–97. https://doi.org/10.1016/j.jneuroim.2005.10.014.
104. Singer HS, Giuliano JD, Hansen BH, Hallett JJ, Laurino JP, Benson M, et al. Antibodies against human putamen in children with Tourette syndrome. Neurology. 1998;50(6):1618–24. https://doi.org/10.1212/wnl.50.6.1618.
105. Kansy JW, Katsovich L, McIver KS, Pick J, Zabriskie JB, Lombroso PJ, et al. Identification of pyruvate kinase as an antigen associated with Tourette syndrome. J Neuroimmunol. 2006;181(1-2):165–76. https://doi.org/10.1016/j.jneuroim.2006.08.007.
106. Harrington LE, Hatton RD, Mangan PR, Turner H, Murphy TL, Murphy KM, et al. Interleukin 17-producing CD4+ effector T cells develop via a lineage distinct from the T helper type 1 and 2 lineages. Nat Immunol. 2005;6(11):1123–32. https://doi.org/10.1038/ni1254.
107. Ruffell B, DeNardo DG, Affara NI, Coussens LM. Lymphocytes in cancer development: polarization towards pro-tumor immunity. Cytokine Growth Factor Rev. 2010;21(1):3–10. https://doi.org/10.1016/j.cytogfr.2009.11.002.
108. Cosmi L, Maggi L, Santarlasci V, Liotta F, Annunziato F. T helper cells plasticity in inflammation. Cytometry A. 2014;85(1):36–42. https://doi.org/10.1002/cyto.a.22348.
109. Eyerich S, Zielinski CE. Defining Th-cell subsets in a classical and tissue-specific manner: examples from the skin. Eur J Immunol. 2014;44(12):3475–83. https://doi.org/10.1002/eji.201444891.
110. Şimşek Ş, Yüksel T, Çim A, Kaya S. Serum cytokine profiles of children with obsessive-compulsive disorder shows the evidence of autoimmunity. Int J Neuropsychopharmacol. 2016;19(8):pyw027. https://doi.org/10.1093/ijnp/pyw027.
111. Swedo SE, Rapoport JL, Cheslow DL, Leonard HL, Ayoub EM, Hosier DM, et al. High prevalence of obsessive-compulsive symptoms in patients with Sydenham's chorea. Am J Psychiatry. 1989;146(2):246–9. https://doi.org/10.1176/ajp.146.2.246.
112. Taylor S. Early versus late onset obsessive-compulsive disorder: evidence for distinct subtypes. Clin Psychol Rev. 2011;31(7):1083–100. https://doi.org/10.1016/j.cpr.2011.06.007.
113. Swedo SE. Pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections (PANDAS). Mol Psychiatry. 2002;7(Suppl 2):S24–5. https://doi.org/10.1038/sj.mp.4001170.

114. Swedo EA, Leckman JF, Rose NR. From research subgroup to clinical syndrome: modifying the PANDAS criteria to describe PANS (pediatric acute-onset neuropsychiatric). Pediatr Therapeut. 2012;2:1–8. https://doi.org/10.4172/2161-0665.1000113.
115. Chiarello F, Spitoni S, Hollander E, Matucci Cerinic M, Pallanti S. An expert opinion on PANDAS/PANS: highlights and controversies. Int J Psychiatry Clin Pract. 2017;21(2):91–8. https://doi.org/10.1080/13651501.2017.1285941.
116. Burchi E, Pallanti S. Antibiotics for PANDAS? Limited evidence: review and putative mechanisms of action. Prim Care Companion CNS Disord. 2018;20(3):17r02232. https://doi.org/10.4088/PCC.17r02232.
117. Marazziti D, Buccianelli B, Palermo S, Parra E, Arone A, Beatino MF, et al. The microbiota/microbiome and the gut-brain axis: how much do they matter in psychiatry? Life (Basel). 2021;11(8):760. https://doi.org/10.3390/life11080760.
118. Turna J, Grosman Kaplan K, Anglin R, Van Ameringen M. "What's bugging the gut in ocd?" A review of the gut microbiome in obsessive-compulsive disorder. Depress Anxiety. 2016;33(3):171–8. https://doi.org/10.1002/da.22454.
119. Rees JC. Obsessive-compulsive disorder and gut microbiota dysregulation. Med Hypotheses. 2014;82(2):163–6. https://doi.org/10.1016/j.mehy.2013.11.026.
120. Dale RC. Tics and Tourette: a clinical, pathophysiological and etiological review. Curr Opin Pediatr. 2017;29(6):665–73. https://doi.org/10.1097/MOP.0000000000000546.
121. Robertson MM, Eapen V, Cavanna AE. The international prevalence, epidemiology, and clinical phenomenology of Tourette syndrome: a cross-cultural perspective. J Psychosom Res. 2009;67(6):475–83. https://doi.org/10.1016/j.jpsychores.2009.07.010.
122. Freeman RD, Fast DK, Burd L, Kerbeshian J, Robertson MM, Sandor P. An international perspective on Tourette syndrome: selected findings from 3,500 individuals in 22 countries. Dev Med Child Neurol. 2000;42(7):436–47. https://doi.org/10.1017/s0012162200000839.
123. Kumar A, Trescher W, Byler D. Tourette syndrome and comorbid neuropsychiatric conditions. Curr Dev Disord Rep. 2016;3(4):217–21. https://doi.org/10.1007/s40474-016-0099-1.
124. Lee WT, Huang HL, Wong LC, Weng WC, Vasylenko T, Jong YJ, et al. Tourette syndrome as an independent risk factor for subsequent sleep disorders in children: a nationwide population-based case-control study. Sleep. 2017;40(3). https://doi.org/10.1093/sleep/zsw072
125. Martino D, Dale RC, Gilbert DL, Giovannoni G, Leckman JF. Immunopathogenic mechanisms in Tourette syndrome: a critical review. Mov Disord. 2009;24(9):1267–79. https://doi.org/10.1002/mds.22504.
126. Landau YE, Steinberg T, Richmand B, Leckman JF, Apter A. Involvement of immunologic and biochemical mechanisms in the pathogenesis of Tourette's syndrome. J Neural Transm (Vienna). 2012;119(5):621–6. https://doi.org/10.1007/s00702-011-0739-x.
127. Abelson JF, Kwan KY, O'Roak BJ, Baek DY, Stillman AA, Morgan TM, et al. Sequence variants in SLITRK1 are associated with Tourette's syndrome. Science. 2005;310(5746):317–20. https://doi.org/10.1126/science.1116502.
128. Worbe Y, Marrakchi-Kacem L, Lecomte S, Valabregue R, Poupon F, Guevara P, et al. Altered structural connectivity of cortico-striato-pallido-thalamic networks in Gilles de la Tourette syndrome. Brain. 2015;138(Pt 2):472–82. https://doi.org/10.1093/brain/awu311.
129. Zapparoli L, Porta M, Paulesu E. The anarchic brain in action: the contribution of task-based fMRI studies to the understanding of Gilles de la Tourette syndrome. Curr Opin Neurol. 2015;28(6):604–11. https://doi.org/10.1097/WCO.0000000000000261.
130. Singer HS, Szymanski S, Giuliano J, Yokoi F, Dogan AS, Brasic JR, et al. Elevated intrasynaptic dopamine release in Tourette's syndrome measured by PET. Am J Psychiatry. 2002;159(8):1329–36. https://doi.org/10.1176/appi.ajp.159.8.1329.
131. Buse J, Schoenefeld K, Münchau A, Roessner V. Neuromodulation in Tourette syndrome: dopamine and beyond. Neurosci Biobehav Rev. 2013;37(6):1069–84. https://doi.org/10.1016/j.neubiorev.2012.10.004.

132. Haugbøl S, Pinborg LH, Regeur L, Hansen ES, Bolwig TG, Nielsen FA, et al. Cerebral 5-HT2A receptor binding is increased in patients with Tourette's syndrome. Int J Neuropsychopharmacol. 2007;10(2):245–52. https://doi.org/10.1017/S1461145706006559.
133. Shaikh N, Leonard E, Martin JM. Prevalence of streptococcal pharyngitis and streptococcal carriage in children: a meta-analysis. Pediatrics. 2010;126(3):e557–64. https://doi.org/10.1542/peds.2009-2648.
134. Martino D, Chiarotti F, Buttiglione M, Cardona F, Creti R, Nardocci N, et al. The relationship between group A streptococcal infections and Tourette syndrome: a study on a large service-based cohort. Dev Med Child Neurol. 2011;53(10):951–7. https://doi.org/10.1111/j.1469-8749.2011.04018.x.
135. Mell LK, Davis RL, Owens D. Association between streptococcal infection and obsessive-compulsive disorder, Tourette's syndrome, and tic disorder. Pediatrics. 2005;116(1):56–60. https://doi.org/10.1542/peds.2004-2058.
136. Murphy TK, Sajid M, Soto O, Shapira N, Edge P, Yang M, et al. Detecting pediatric autoimmune neuropsychiatric disorders associated with streptococcus in children with obsessive-compulsive disorder and tics. Biol Psychiatry. 2004;55(1):61–8. https://doi.org/10.1016/s0006-3223(03)00704-2.
137. Müller N, Riedel M, Förderreuther S, Blendinger C, Abele-Horn M. Tourette's syndrome and mycoplasma pneumoniae infection. Am J Psychiatry. 2000;157(3):481–2. https://doi.org/10.1176/appi.ajp.157.3.481-a.
138. Ercan TE, Ercan G, Severge B, Arpaozu M, Karasu G. Mycoplasma pneumoniae infection and obsessive-compulsive disease: a case report. J Child Neurol. 2008;23(3):338–40. https://doi.org/10.1177/0883073807308714.
139. Riedel M, Straube A, Schwarz MJ, Wilske B, Müller N. Lyme disease presenting as Tourette's syndrome. Lancet. 1998;351(9100):418–9. https://doi.org/10.1016/S0140-6736(05)78357-4.
140. Krause D, Matz J, Weidinger E, Wildenauer A, Obermeier M, Riedel M, et al. Association between intracellular infectious agents and Tourette's syndrome. Eur Arch Psychiatry Clin Neurosci. 2010;260(4):359–63. https://doi.org/10.1007/s00406-009-0084-3.
141. Matz J, Krause DL, Dehning S, Riedel M, Gruber R, Schwarz MJ, et al. Altered monocyte activation markers in Tourette's syndrome: a case-control study. BMC Psychiatry. 2012;12:29. https://doi.org/10.1186/1471-244X-12-29.
142. Müller N. Anti-inflammatory therapy with a COX-2 inhibitor in Tourette's syndrome. Inflammopharmacology. 2004;12(3):271–5. https://doi.org/10.1163/1568560042342338.
143. Cohen B, Preuss CV. Celecoxib. In: StatPearls [Internet]. Treasure Island, FL: StatPearls Publishing; 2021.
144. Huys D, Hardenacke K, Poppe P, Bartsch C, Baskin B, Kuhn J. Update on the role of antipsychotics in the treatment of Tourette syndrome. Neuropsychiatr Dis Treat. 2012;8:95–104. https://doi.org/10.2147/NDT.S12990.
145. Pringsheim T, Okun MS, Müller-Vahl K, Martino D, Jankovic J, Cavanna AE, et al. Practice guideline recommendations summary: treatment of tics in people with Tourette syndrome and chronic tic disorders. Neurology. 2019;92(19):896–906. https://doi.org/10.1212/WNL.0000000000007466.
146. Singer HS, Wong DF, Brown JE, Brandt J, Krafft L, Shaya E, et al. Positron emission tomography evaluation of dopamine D-2 receptors in adults with Tourette syndrome. Adv Neurol. 1992;58:233–9.
147. Gunther J, Tian Y, Stamova B, Lit L, Corbett B, Ander B, et al. Catecholamine-related gene expression in blood correlates with tic severity in Tourette syndrome. Psychiatry Res. 2012;200(2-3):593–601. https://doi.org/10.1016/j.psychres.2012.04.034.
148. Kipnis J, Cardon M, Avidan H, Lewitus GM, Mordechay S, Rolls A, et al. Dopamine, through the extracellular signal-regulated kinase pathway, downregulates CD4+CD25+ regulatory T-cell activity: implications for neurodegeneration. J Neurosci. 2004;24(27):6133–43. https://doi.org/10.1523/JNEUROSCI.0600-04.2004.

149. Tian Y, Gunther JR, Liao IH, Liu D, Ander BP, Stamova BS, et al. GABA- and acetylcholine-related gene expression in blood correlate with tic severity and microarray evidence for alternative splicing in Tourette syndrome: a pilot study. Brain Res. 2011;1381:228–36. https://doi.org/10.1016/j.brainres.2011.01.026.
150. Zhou FM, Wilson CJ, Dani JA. Cholinergic interneuron characteristics and nicotinic properties in the striatum. J Neurobiol. 2002;53(4):590–605. https://doi.org/10.1002/neu.10150.
151. Rane MJ, Gozal D, Butt W, Gozal E, Pierce WM Jr, Guo SZ, et al. γ-amino butyric acid type B receptors stimulate neutrophil chemotaxis during ischemia-reperfusion. J Immunol. 2005;174(11):7242–9. https://doi.org/10.4049/jimmunol.174.11.7242.
152. Lennington JB, Coppola G, Kataoka-Sasaki Y, Fernandez TV, Palejev D, Li Y, et al. Transcriptome analysis of the human striatum in Tourette syndrome. Biol Psychiatry. 2016;79(5):372–82. https://doi.org/10.1016/j.biopsych.2014.07.018.
153. Kumar A, Williams MT, Chugani HT. Evaluation of basal ganglia and thalamic inflammation in children with pediatric autoimmune neuropsychiatric disorders associated with streptococcal infection and Tourette syndrome: a positron emission tomographic (PET) study using 11C-[R]-PK11195. J Child Neurol. 2015;30(6):749–56. https://doi.org/10.1177/0883073814543303.
154. Ueno M, Fujita Y, Tanaka T, Nakamura Y, Kikuta J, Ishii M, et al. Layer V cortical neurons require microglial support for survival during postnatal development. Nat Neurosci. 2013;16(5):543–51. https://doi.org/10.1038/nn.3358.
155. Borish LC, Steinke JW. 2. Cytokines and chemokines. J Allergy Clin Immunol. 2003;111(2 Suppl):S460–75. https://doi.org/10.1067/mai.2003.108.
156. Yeon SM, Lee JH, Kang D, Bae H, Lee KY, Jin S, Kim JR, et al. A cytokine study of pediatric Tourette's disorder without obsessive-compulsive disorder. Psychiatry Res. 2017;247:90–6. https://doi.org/10.1016/j.psychres.2016.11.005.
157. Gabbay V, Coffey BJ, Guttman LE, Gottlieb L, Katz Y, Babb JS, et al. A cytokine study in children and adolescents with Tourette's disorder. Prog Neuropsychopharmacol Biol Psychiatry. 2009;33(6):967–71. https://doi.org/10.1016/j.pnpbp.2009.05.001.
158. Brambilla F, Perna G, Bellodi L, Arancio C, Bertani A, Perini G, et al. Plasma interleukin-1β and tumor necrosis factor concentrations in obsessive-compulsive disorders. Biol Psychiatry. 1997;42(11):976–81. https://doi.org/10.1016/s0006-3223(96)00495-7.
159. Pingle SK, Tumane RG, Jawade AA. Neopterin: biomarker of cell-mediated immunity and potent usage as biomarker in silicosis and other occupational diseases. Indian J Occup Environ Med. 2008;12(3):107–11. https://doi.org/10.4103/0019-5278.44690.
160. Hoekstra PJ, Anderson GM, Troost PW, Kallenberg CG, Minderaa RB. Plasma kynurenine and related measures in tic disorder patients. Eur Child Adolesc Psychiatry. 2007;16(Suppl 1):71–7. https://doi.org/10.1007/s00787-007-1009-1; Erratum in: Eur Child Adolesc Psychiatry. 2007 Dec;16(8):537
161. Yildirim Z, Karabekiroglu K, Yildiran A, Celiksoy MH, Artukoglu B, Baykal S, et al. An examination of the relationship between regulatory T cells and symptom flare-ups in children and adolescents diagnosed with chronic tic disorder and Tourette syndrome. Nord J Psychiatry. 2021;75(1):18–24. https://doi.org/10.1080/08039488.2020.1779808.
162. Dominguez-Villar M. Hafler DA (2018) Regulatory T cells in autoimmune disease. Nat Immunol. 2018;19(7):665–73. https://doi.org/10.1038/s41590-018-0120-4.
163. Kawikova I, Leckman JF, Kronig H, Katsovich L, Bessen DE, Ghebremichael M, et al. Decreased numbers of regulatory T cells suggest impaired immune tolerance in children with Tourette syndrome: a preliminary study. Biol Psychiatry. 2007;61(3):273–8. https://doi.org/10.1016/j.biopsych.2006.06.012.
164. Ferrari M, Termine C, Franciotta D, Castiglioni E, Pagani A, Lanzi G, et al. Dopaminergic receptor D5 mRNA expression is increased in circulating lymphocytes of Tourette syndrome patients. J Psychiatr Res. 2008;43(1):24–9. https://doi.org/10.1016/j.jpsychires.2008.01.014.

165. Möller JC, Tackenberg B, Heinzel-Gutenbrunner M, Burmester R, Oertel WH, Bandmann O, et al. Immunophenotyping in Tourette syndrome – a pilot study. Eur J Neurol. 2008;15(7): 749–53. https://doi.org/10.1111/j.1468-1331.2008.02159.x.
166. Marzio R, Mauël J, Betz-Corradin S. CD69 and regulation of the immune function. Immunopharmacol Immunotoxicol. 1999;21(3):565–82. https://doi.org/10.3109/08923979909007126.
167. Green DR, Droin N, Pinkoski M. Activation-induced cell death in T cells. Immunol Rev. 2003;193:70–81. https://doi.org/10.1034/j.1600-065X.2003.00051.x.
168. Pranzatelli MR, Tate ED, Allison TJ. Case-control, exploratory study of cerebrospinal fluid chemokines/cytokines and lymphocyte subsets in childhood Tourette syndrome with positive streptococcal markers. Cytokine. 2017;96:49–53. https://doi.org/10.1016/j.cyto.2017.03.003.
169. Perlmutter SJ, Leitman SF, Garvey MA, Hamburger S, Feldman E, Leonard HL, et al. Therapeutic plasma exchange and intravenous immunoglobulin for obsessive-compulsive disorder and tic disorders in childhood. Lancet. 1999;354(9185):1153–8. https://doi.org/10.1016/S0140-6736(98)12297-3.
170. Murphy TK, Parker-Athill EC, Lewin AB, Storch EA, Mutch PJ. Cefdinir for recent-onset pediatric neuropsychiatric disorders: a pilot randomized trial. J Child Adolesc Psychopharmacol. 2015;25(1):57–64. https://doi.org/10.1089/cap.2014.0010.
171. Shalbafan M, Mohammadinejad P, Shariat SV, Alavi K, Zeinoddini A, Salehi M, et al. Celecoxib as an adjuvant to fluvoxamine in moderate to severe obsessive-compulsive disorder: a double-blind, placebo-controlled, randomized trial. Pharmacopsychiatry. 2015;48(4-5): 136–40. https://doi.org/10.1055/s-0035-1549929.

# Molecular Imaging of Neuroinflammation in Alzheimer's Disease and Mild Cognitive Impairment

14

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**Abstract**

Alzheimer's disease (AD) is the most prevalent neurocognitive disorder. Due to the ineffectiveness of treatments targeting the amyloid cascade, molecular biomarkers for neuroinflammation are attracting attention with increasing knowledge about the role of neuroinflammation in the pathogenesis of AD. This chapter will explore the results of studies using molecular imaging for diagnosing AD and mild cognitive impairment (MCI). Because it is critical to interpreting the data to understand which substances are targeted in molecular imaging, this chapter will discuss the two most significant targets, microglia and astrocytes, as well as the best-known radioligands for each. Then, neuroimaging results with PET neuroinflammation imaging will be reviewed for AD and MCI. Although a growing body of evidence has suggested that these molecular imaging biomarkers for neuroinflammation may have a role in the diagnosis of AD and MCI, the findings are inconsistent or cross-sectional, which indicates that it is difficult to apply the contents in practice due to the need for additional study. In particular, because the results of multiple interventions targeting neuroinflammation were inconclusive, molecular imaging markers for neuroinflammation can be used in combination with conventional markers to

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select appropriate patients for early intervention for neuroinflammation rather than as a single marker.

**Keywords**

Alzheimer's disease · Mild cognitive impairment · Neuroinflammation · Biomarkers · Neuroimaging · Microglia · Astrocytes

## 14.1 Introduction

Alzheimer's disease (AD) is the most prevalent major neurocognitive disorder in the world [1], and its incidence among the elderly is increasing at an alarming rate [2, 3]. Around 10% of people aged 65 and over are considered to have AD [4]; this proportion rises to 32% in those aged 85 and more, with an annual incidence of 6.48% [5]. Individuals diagnosed with AD experience a reduction in their quality of life and impairment over time, which finally results in death [6]. A further point to mention is that in 2015, the average annual socioeconomic cost per patient was $19,144.36, with total costs of $167.74 billion. According to projections, overall annual expenses are estimated to reach $507.49 billion in 2030, increasing to $1.89 trillion in 2050 [7]. Atri et al. state that early and accurate diagnosis is crucial for optimal treatment [8], but this is challenging since the current diagnostic approach for AD relies on clinical observation and objective neuropsychiatric testing [9].

Amyloid-β (Aβ) plaques and abnormal tau tangles in the brain are pathological markers of AD [10]. The amyloid cascade hypothesis, which asserts that Aβ accumulation initiates a cascade of events that end in neuronal destruction, was proposed in light of the historical evolution of amyloid and tau diseases and evidence that Aβ overproduction results in AD [11]. However, numerous treatments targeting Aβ plaques and neurofibrillary tangles are limited in their ability to modify the course of AD [12]; thus, additional AD pathologies for intervention have been proposed [13]. In particular, neuroinflammation is thought to be a key pathogenic characteristic of AD [14]. These results highlight the critical need for accurate in vivo neuroinflammation assessment, allowing researchers to understand better the neuroimmune processes that contribute to neurological illness and better guide clinical trial design [15].

Unlike in autoimmune disorders of the central nervous system (CNS) such as multiple sclerosis, in which CNS antigen-specific T cells infiltrate the brain and spinal cord [16], neuroinflammation in AD originates in the brain and begins near the Aβ plaques and involves inflammatory activation [17]. The accumulation of Aβ plaques is thought to be a major component driving the neuroinflammatory response in AD associated with microglia activation [17–19]. Activated microglia are consistently seen in proximity to Aβ plaques in postmortem immunohistochemical investigations of AD patients' brain slices [20, 21]. Microglial cells may bind to soluble and fibrillar Aβ through cell surface receptors causing inactivation and

cytokine production [22]. Aβ clearance by microglia via receptor-mediated phagocytosis and degradation has also been demonstrated in vitro [23, 24].

There are several methods for detecting neuroinflammation, but neuroimaging is important in terms of being able to examine the degree of inflammation in the brain. Positron emission tomography (PET) scanning may be a valuable tool in the study of neuroinflammation by enabling researchers to elucidate the interplay between inflammatory processes and neurodegenerative disorders and allow for the early detection of novel treatment strategies. PET scanning provides us with the ability to identify, measure, and define the morphology of the brain's inflammatory response [16]. This chapter focuses on results from molecular imaging studies using PET in AD and mild cognitive impairment (MCI) that investigated neuroinflammation.

## 14.2 Positron Emission Tomography (PET)

The advent of numerous noninvasive imaging techniques has significantly aided our understanding of the brain's architecture and function. PET is a type of functional imaging that allows for the in vivo observation and quantification of metabolic processes in mammalian biology [25]. PET is particularly well suited for assessing neuroinflammation and has the ability to discriminate between components of the neuroimmune response due to its power to identify specific proteins at low levels [15]. To obtain PET images, a ligand must be developed with a high degree of specificity for a single target and a low degree of nonspecific binding [26]. Particles of positron-emitting radioisotopes are attached to the molecular probe or ligand (e.g., $^{18}F$, $^{11}C$, $^{15}O$). The radioligand is then administered intravenously at a tracer dose to ensure that it occupies the fewest possible target locations in the brain (generally defined as less than 5% of the total accessible target in the brain). After being injected intravenously, the radioligand decays, generating positrons (+) from its nucleus as it degrades. When one electron collides with another electron in the tissue, annihilation happens, converting their masses to their energy counterparts through the emission of two 511-keV photons that are 180° apart [25]. Scintillation detectors positioned around the participant detect the photons, allowing for calculation of the radioligand's spatial distribution in the brain and, therefore, the analysis of the biological process under research.

## 14.3 Targets of PET Concerning Neuroinflammation in AD and MCI

### 14.3.1 Microglia

A type of myeloid lineage immune cell called microglia is found in the CNS. Microglial cells, which are mononuclear phagocytes found throughout the brain and account for about 10% of the total cell population in the CNS, serve as the first line of defense against invading pathogens and other harmful chemicals [27, 28]. In

the resting state, microglial cells have a ramified shape. It is via microglial receptors that danger signals are detected, and this is what triggers microglial activation. The activated state is associated with a surge in the number of resident microglia and morphological and physiological alterations in the cells, all of which lead to an amoeboid morphology and increase expression of major histocompatibility complex II [29].

Genome-wide association studies have identified variants in genes associated with innate immunity as risk factors for AD, underscoring the crucial role of microglia in the genesis and progression of AD responding to neurodegeneration [30]. Microglial activity is a neuropathological hallmark of AD in humans, and microglia contribute to the development of neuritic plaques by concentrating near Aβ deposits [31]. Activated microglia seem to develop after Aβ plaques but before tau pathology, according to a mathematical model [32]. However, longitudinal PET imaging experiments with ligands for detecting microglia, Aβ, and tau are required to investigate the nature of these processes in vivo.

### 14.3.2 Astrocytes

Astrocytes are specialized glial cells that serve as the scaffolding for the whole central nervous system (CNS). They can be protoplasmic (found chiefly in grey matter) or fibrous (found primarily in white matter) [33]. Along with endothelial cells and tightly enclosed synapses, astrocyte processes contribute to the blood-brain barrier (BBB). As predicted by their anatomical niche, astrocytes regulate cerebral blood flow, maintain fluid and neurotransmitter balance, induce synaptic development, and provide metabolic and neurotrophic support for synapses [34–36]. Additionally, astrocytes generate distinct perivascular channels in the central nervous system, referred to as the glymphatic system, which removes neurotoxic molecules such as Aβ and tau tangles [37, 38].

Astrocytes contribute to neuroinflammatory processes through their response to various pro- and anti-inflammatory factors [39]. Microglia and astrocytes both have a role in the phagocytosis of cell debris and A, as well as in response to injury [40]. Additionally, as demonstrated in preclinical investigations, astrocytes contribute to neuronal metabolic support [41]. A increases glucose absorption in primary cultures of astrocytes [42], changing their metabolic profile. As a consequence, monoamine oxidase B (MAO-B) expression is increased in astrocytes [43] and is a potential molecular target for imaging astrocytes. Overexpression of astrocytic MAO-B resulted in elevated levels of gamma-aminobutyric acid (GABA) and neurotoxic glutamate in a transgenic mouse model of AD, interrupting homeostasis and resulting in cognitive deficits [44].

Astrocytes seem to attempt to reestablish homeostasis during the early stages of AD with their multiple housekeeping functions [42]. Astrocytes have also been shown to have amyloid-containing granules in the region of plaques in human brains [45], indicating an effort by astrocytes to remove amyloid accumulations throughout the illness process [46]. Additionally, studies have shown that astrocytes move

toward Aβ plaques and destroy Aβ both in vitro and in vivo [47, 48]. Reactive astrocytes were discovered to generate GABA and glutamate excessively in an animal model of AD, resulting in poor memory and synaptic loss. Additionally, these cells disrupted both microcirculation and BBB, which increase A deposition and disease development. Reactive astrocytes may possibly pave the path for the initial amyloid plaques to develop. Notably, astrocytes collaborate closely with microglial cells and may mediate some of microglia's harmful effects during disease states.

## 14.4 Targets for Detecting Microglial Activity in AD and MCI Patients

### 14.4.1 18-kDa Translocator Protein (TSPO)

An increase in or de novo production of a number of cell-surface and cytoplasmic substances is seen in microglia that have been activated [49]. Translocator protein 18-kDa (TSPO) is one protein that has piqued researchers' attention for measuring neuroinflammation in vivo. TSPO was discovered through investigations of central benzodiazepine receptor (CBR) binding [50]. It was first referred to as a peripheral benzodiazepine receptor (PBR) due to its widespread distribution in peripheral organs [51–53].

A variety of physiological functions, including cellular respiration, cholesterol transport, and immunomodulation, are regulated by TSPO expression [54]. The voltage-dependent anion channel and the adenine nucleotide carrier, both of which are 30 kDa, may form a multimeric complex in the outer mitochondrial membrane [55, 56]. While TSPO is distributed throughout the cell, the outer mitochondrial membrane is the main intracellular site [57]. However, the precise physiological activities of TSPO remain unknown. Despite this, it is thought to be involved in a range of activities, including cell growth, bile acid production, steroidogenesis, cell metabolism, cholesterol transport, calcium flow, apoptosis, and neuroinflammation [58, 59].

TSPO is found at low levels throughout the normal central nervous system, including endothelial cells, the epidermis, the choroid plexus, the olfactory bulb, and particular sparse glial cells [60, 61]. In response to brain injury, normal aging, and illnesses of the CNS such as AD, cerebrovascular disease, and multiple sclerosis, this expression increases rapidly from a relatively low baseline level [53, 59, 62]. Microautoradiography and immunohistochemical investigations have shown that regions with elevated TSPO levels also exhibit an increase in microglia [63, 64] and have connected this upregulation to microglial cell activation.

TSPO–radioligand binding was found to be correlated with the number of activated microglia in postmortem tissues from individuals suffering from a variety of neurological diseases [65]. This finding indicated that TSPO may be associated with microglial proliferation, migration, and phagocytic capacity [56]. Moreover, it's worth noting that reactive astrocytes have been shown to have higher TSPO

expression [66, 67]. Because TSPO levels are low in the brain parenchyma in the normal state and rise regionally in response to brain injuries, it is an excellent marker for molecular imaging for neuroinflammation.

### 14.4.2 Existing TSPO Radioligands

Numerous TSPO radioligands have been produced throughout the years, some of which have been investigated in human populations in vivo. [$^{11}$C]-PK11195, a specific antagonist for TSPO, is the most frequently utilized radioligand. [$^{11}$C]-PK11195 was initially utilized as a racemic mixture [66]. However, because more recent research discovered that [$^{11}$C]-(R)-PK11195, the R-enantiomer of [$^{11}$C]-PK11195, had higher affinity for TSPO than the S-enantiomer [68], [$^{11}$C]-(R)-PK11195 has recently been employed to study neuroinflammation. [$^{11}$C]-PK11195 has been utilized in the diagnosis of various neurologic diseases including AD [69], Parkinson's disease [70], multiple sclerosis [71], cerebrovascular disease [72], and Huntington's disease [70].

However, there are some technological constraints, and the inherent characteristics of the chemical associated with [$^{11}$C]-PK11195 have hampered utilizing this for investigating neuroinflammation in clinic. First, carbon-11 radiolabeling is a complex process, and the half-life of carbon-11 is just 20 min long; thus, [$^{11}$C]-PK11195 could only be used in PET centers equipped with cyclotrons [16]. Second, [$^{11}$C]-PK11195 has a low signal-to-noise ratio because of its nonspecific binding associated with high lipophilicity, low BBB penetration, high plasma binding, and low bioavailability [73]. As a result, it is limited in its capacity to detect minor changes in TSPO expression [74]. Because of the limitations of [$^{11}$C]-PK11195, a lot of research has gone into developing better radioligands.

Newly developed second-generation TSPO ligands have presented greater affinity for TSPO and improved kinetic properties. Only those that have been researched extensively in humans, particularly in AD, will be included in this chapter. In the monkey brain, [$^{11}$C]-PBR28 showed a stronger specific signal for microglial activation than [$^{11}$C]-PK11195 due to higher affinity, higher BBB penetration, and more specific binding with lower lipophilicity [75, 76]. In healthy brain research, [$^{11}$C]-DPA-713 was shown to be more sensitive than [$^{11}$C]-PK11195 for detecting enhanced expression of TSPO [77]. This claim was supported by recent research, which found that [$^{11}$C]-DPA-713 shows higher TSPO density in more broad brain areas of aging individuals and AD patients than [$^{11}$C]-PK11195 [78]. The ligand [$^{11}$C]-DAA1106 has been shown to bind to activated microglial cells in CNS diseases with higher affinity (tenfold greater than [$^{11}$C]-PK11195), resulting in greater contrast between lesioned and unlesioned regions [59, 65, 79, 80].

The discovery of novel chemicals radiolabeled with [$^{18}$F] that showed higher affinity, stronger specific signal, higher bioavailability, and longer half-life than [$^{11}$C] has permitted PET centers without on-site cyclotrons to utilize PET for detecting neuroinflammation. Preclinical investigations in nonhuman primates have revealed that [$^{18}$F]-FEDAA1106 has a stronger TSPO affinity than

[$^{11}$C]-PK11195 and [$^{11}$C]-DAA1106 and that [$^{18}$F]-FEDAA1106 has a higher BBB penetration than [$^{11}$C]-PK11195 and [$^{11}$C]-DAA1106 [81, 82]. It's also been claimed that [$^{18}$F]-FEMPA is a good tracer for TSPO [83]. [$^{18}$F]-FEPPA had a high TSPO affinity and good BBB penetration and pharmacokinetics [84]. Vinpocetine is a neuroprotective compound that may potentially have anti-inflammatory effects. Radiolabeled vinpocetine has also been shown to be a potential TSPO marker [85]. Gulyás et al. [86] suggested that its good brain penetration compensates for a low TSPO affinity [86].

### 14.4.3 Genetic Polymorphism Affecting TSPO Quantification by PET

In comparison with [$^{11}$C]-PK11195, second-generation TSPO radioligands exhibit greater affinity and brain uptake and a higher signal-to-noise ratio. However, second-generation TSPO radiotracers are limited in their sensitivity to a polymorphism in the TSPO gene that causes an alanine-to-threonine substitution [87]. Due to this polymorphism, these ligands have a varied affinity for TSPO, resulting in three distinct binding patterns [88, 89]. These patterns are high affinity binders (HAB) and low affinity binders (LAB), which are homozygotes that express Ala or Thr, respectively, and mixed affinity binders (MAB), which are heterozygotes that express both Ala and Thr [87, 90]. Because the TSPO polymorphism impacts the binding of all second-generation radioligands, participants must be genotyped to allow for precise measurement of TSPO availability [91]. Notably, there were no significant differences among the three TSPO affinity subgroups of individuals with AD when examining clinical characteristics, amyloid deposition, and degree of cognitive impairment [92, 93]. These results reduce the possibility of bias that may occur when the interpretation of PET imaging results obtained from the TSPO subgroup is applied to the AD population.

### 14.4.4 Other Radiotracers Targeting Microglial Activation

This section presents alternative radioligands for active imaging microglia that do not use TSPO as a target. Additional research is needed to find new targets related to microglia migratory or phagocytic capabilities more than to investigate upregulated proteins in active microglial cells. These targets could be identified and prioritized for future exploration utilizing innovative methodologies such as cell type-specific transcriptional profiling, which uncovered multiple cell type-specific alterations previously unreported in whole tissue RNA [94].

Immune cells such as monocytes and macrophages express the cannabinoid type 2 receptor (CB2R), and CB2R is largely present on microglia in the brain [95]. The CB2R is a component of the endogenous cannabinoid system and serves as an alternate membrane signal for microglial activation, resulting in increased expression. Although its upregulation has been reported in an AD animal model, an in vivo study that evaluated [$^{11}$C]-NE40—a tracer for CB2R—in healthy controls and

patients with AD presented decreased CB2R availability in vivo in AD patients compared with preclinical and postmortem data [96]. This discrepancy is most likely caused by the extremely low level of CB2R expression and an inadequate selectivity for CB2R; thus additional CB2R agonists with high affinity are being developed [97].

Ketoprofen is a selective COX-1 inhibitor, which is found in high concentrations in activated microglia [98]. Ketoprofen pro-radiotracer ([$^{11}$C]-KTP) enhances the drug's BBB penetration [99]. Animal studies have shown that [$^{11}$C]-KTP is maintained in inflammatory lesions due to the presence of COX-1 in the tissues. According to the results of the first in vivo research conducted on a healthy population, [$^{11}$C]-KTP showed characteristics as a stable and safe PET tracer with high BBB penetration [100]. Despite this, the washout rate was not significantly different in AD patients compared with controls, indicating that further research and development are needed before this test can be regarded as a viable neuroinflammation indicator [101].

Moreover, radioligands related to different targets are being developed. Studies using radioligands that detect the nicotinic acetylcholine receptors have also reported significant AD-related results. Recently, 2-[$^{18}$F]-fluoro-A85380, a radioligand for nicotinic acetylcholine receptors, was found to be upregulated to the same extent as TSPO in activated microglia and astrocytes compared with [$^{11}$C]-PK11195 [102]. Additionally, recent studies using novel radioligands for nicotinic acetylcholine receptors, such as [$^{18}$F]-ASEM and [$^{18}$F]-DBT-10, showed encouraging results with varying degrees of effectiveness [103–105].

## 14.5 Targets for Detecting Activity of Astrocytes in AD and MCI Patients

Numerous molecular imaging markers are required to investigate astrocytosis since it is a highly dynamic process that sequentially progresses from protective to harmful phases [106]. However, there is mounting evidence that morphology and function are linked in astrocyte activation, which can be characterized by increased expression of MAO-B and intermediate filaments including nestin, vimentin, and glial fibrillary acidic protein (GFAP) [107].

### 14.5.1 Enzymes

MAO-B expression is increased in reactive astrocytes during neuroinflammatory processes. L-deprenyl is a highly specific irreversible inhibitor of the MAO-B enzyme. The radioactive isotope [$^{11}$C]-L-deprenyl ([$^{11}$C]-DED) has been used to study the distribution of MAO-B in the brain and, on occasion, to evaluate the degree to which other MAO-B inhibitors bind to it [108, 109]. Increased regional binding was shown to correlate with an increased number of activated astrocytes in Alzheimer brains in a postmortem investigation [110]. Autoradiography

investigations have shown that the binding of 3H-L-deprenyl partially coincides with that of GFAP in AD and other neurodegenerative disorders [111], suggesting that MAO-B has a high degree of selectivity for activated astrocytes. The most significant absorption of [$^{11}$C]-L-deprenyl in AD brain tissue was seen during the first Braak stages, suggesting an early involvement of astrocytosis in AD [110].

[$^{11}$C]-DED exhibited favorable kinetics as a radioligand, and its binding is not reliant on brain perfusion [112]. PET imaging using [$^{11}$C]-DED has been utilized to study astrocytosis in neurodegenerative disorders such as AD [113, 114]. Multitracer PET scans using [$^{11}$C]-DED, [$^{11}$C]-PIB, and [$^{18}$F]-fluorodeoxyglucose ([$^{18}$F]-FDG) enabled the investigation of the spatiotemporal patterns of astrocytosis, fibrillar Aβ deposition, and glucose metabolism at various phases of illness development. In these investigations, prodromal AD was shown to have substantially higher [$^{11}$C]-DED binding than healthy controls or AD patients [114]. Astrocytosis was detected early in the presymptomatic phases of autosomal-dominant AD using [$^{11}$C]-DED PET [48]; longitudinally, A plaque deposition ([$^{11}$C]-PIB) increased while astrocytosis ([$^{11}$C]-DED) decreased [115].

### 14.5.2 Other Markers

Historically, [$^{18}$F]-FDG-PET hypometabolism was considered a biomarker for neurodegeneration and neuronal injury. However, the previous study found that activating astrocytes resulted in extensive graded glucose absorption in rodent brain using [$^{18}$F]-FDG-PET [116]. This research provides further evidence for the astrocyte-neuron lactate shuttle theory, which was proposed 20 years ago and stated that most neuronal energy requirements are supplied by lactate produced in astrocytes and shuttled to neurons [117, 118]. Consistent with these preclinical results, a longitudinal decrease in astrocytosis, as assessed by MAO-B expression, was recently found to be associated with progressive hypometabolism in autosomal-dominant AD mutation carriers [119], suggesting that astrocytes represent metabolic activity in AD. The observed decrease in MAO-B, which may indicate decreased astrocyte glucose demand, may represent neurodegeneration by astrocytes, a phenotype associated with late-stage AD [107]. This association must be verified in research comparing [$^{18}$F]-FDG-PET imaging of AD patients with the postmortem examination of humans.

Other possible targets have been investigated in preclinical research, although in vivo human trials remain uncommon. Astrocyte-specific glutamate transporters GLT1 (in rats) and EAAT2 (in humans) were decreased in postmortem tissue [120, 121], indicating that astrocytes lose function throughout late illness stages. Similarly, glutamine synthetase expression decreased with age in a transgenic mouse model of AD [122]. Another possible indicator of astrocyte-associated metabolic failure is a decrease in the expression of the GLUT1 (glucose transporter 1) protein, a glucose transporter that is primarily expressed in astrocytes [123]. Interestingly, aerobic glycolysis, which is believed to occur mainly in astrocytes, was shown to decrease as tau accumulated in preclinical AD patients [124], indicating that

**Table 14.1** A list of existing radioligands for detecting neuroinflammation of AD and MCI concerning targets

| Targets | | Existing radioligands | | | |
|---|---|---|---|---|---|
| Microglia | 18-kDa translocator protein | [$^{11}$C]-PK11195 | [$^{11}$C]-PBR28 | [$^{11}$C]-DPA-713 | [$^{11}$C]-DAA1106 |
| | | [$^{18}$F]-FEMPA | [$^{18}$F]-FEDAA1106 | [$^{18}$F]-PBR06 | [$^{18}$F]-DPA714 |
| | Cannabinoid type 2 receptor | [$^{11}$C]-NE40 | | | |
| | Ketoprofen | [$^{11}$C]-KTP-Me | | | |
| | Nicotinic acetylcholine receptors | 2-[$^{18}$F]-fluoro-A85380 | | [$^{18}$F]-ASEM | [$^{18}$F]-DBT-10 |
| Astrocyte | Monoamine oxidase B | [$^{11}$C]-DED | | | |
| | Metabolic markers | [$^{18}$F]-FDG | | | |
| | Adenosine A2A receptors | [$^{11}$C]-TMSX | | | |

astrocyte dysfunction occurs early in AD. These findings provide the impetus for ongoing research into PET imaging tracers that may specifically target astrocyte-specific glutamate transporters in the human brain, such as EAAT1/EAAT2 and GLAST (glutamate aspartate transporter). These investigations will significantly advance our knowledge of how astrocytes contribute to the metabolic alterations seen in AD.

Adenosine binding to adenosine A2A receptors (A2AR) has been shown to reduce inflammation, resulting in an increase in the expression of A2AR in areas of neuroinflammation and tissue injury to control the endogenous inflammation. [$^{11}$C]-TMSX, a radioligand for adenosine A2ARs, has been utilized in vivo to investigate healthy controls, individuals with Parkinson's disease, and those with multiple sclerosis (Table 14.1) [125, 126].

## 14.6 PET Imaging of Neuroinflammation in AD

[$^{11}$C]-PK11195, has been used in various studies with contradictory results. In two studies, increased uptake of [$^{11}$C]-PK11195 was reported in AD patients: using SPECT in the frontal and mesotemporal regions [127] and using region-of-interest analysis in the frontal, parietal, temporal, cingulate cortices, and occipital as well as the striatum [128]. Another early research using [$^{11}$C]-PK11195 demonstrated a significant increase not only in binding across multiple cortical areas but also in regions that are generally unaffected in the early stages of AD such as the cerebellum and striatum [69]. These findings were replicated in the other study that demonstrated an increase in cortical [$^{11}$C]-PK11195 binding that remained significant in the frontal cortex following multiple comparisons correction in AD patients

with positive [$^{11}$C]-PIB-PET imaging [129]. However, another initial investigation of AD patients using [$^{11}$C]-PK11195 failed to elucidate TSPO binding sites linked with microglial activation in dementia patients [130]. Wiley et al. [131] suggested that microglial activation is either not detectable in mild to moderate AD or is restricted to the latter stages of severe AD [131]. Recent research using voxel-wise statistical parametric mapping (SPM) analysis of [$^{11}$C]-PK11195 revealed inconsistent findings; one study indicated enhanced binding [132], while one found no change between diagnostic groups [133]. Interestingly, another longitudinal study using [$^{11}$C]-PK11195 discovered distinct patterns of microglial activation between MCI and AD: the AD group demonstrated an increase in binding over time, while in contrast, the MCI group demonstrated decreasing levels of binding over time [134].

The second generation of TSPO ligands enabled the continuation of PET studies in AD with advanced specificity. However, the conclusions of studies that do not account for the genetic status of TSPO binding (as discussed in "Genetic polymorphism affecting TSPO quantification by PET") must be interpreted cautiously. Numerous studies using second-generation TSPO ligands discovered that AD patients had increased binding compared with controls. The neuroanatomical areas associated with increased binding have varied across studies. Most studies presented widespread cortical binding while some also demonstrated increases in specific brain regions: the frontal, temporal, and parietal cortex ([$^{18}$F]-DPA-714) [93]; the parietal and temporal cortices including hippocampus, entorhinal cortex, precuneus, and occipital cortex ([$^{11}$C]-PBR28) [135–137]; and the medial and lateral temporal cortex, posterior cingulate, caudate, putamen, and thalamus ([$^{18}$F]-FEMPA) [83]. Only one study has examined binding in the white matter [138] and reported a significant increase in the cingulum bundle and posterior limb of the internal capsule.

### 14.6.1 Subtypes of AD

Additionally, several PET investigations have examined the connection between neuroinflammation and subtypes of AD such as age of onset and AD variants. Concerning clinical features of AD, the age of onset was shown to have an impact in the study using [$^{11}$C]-PBR28, with early-onset AD (<65 years) patients showing higher TSPO binding than late-onset AD patients [135]. In contrast, no connection with onset age was reported in studies utilizing [$^{18}$F]-DPA-714 on a larger cohort of individuals [93]. Moreover, more recent research using [$^{18}$F]-FEPPA found no link between TSPO binding and disease severity or duration of illness [138].

Even though the importance of AD variants has been emphasized in AD dementia, only a few studies have examined putative differences in TSPO binding patterns among AD types. Kreisl et al. [139] demonstrated increased [$^{11}$C]-PBR28 binding in posterior cortical atrophy-PCA areas of the occipital, posterior parietal, and temporal lobes, most notably with a pattern of hypometabolism shown by fluorodeoxyglucose (FDG) PET imaging in AD [140]. In comparison, individuals with amnestic AD patients had much higher [$^{11}$C]-PBR28 binding in the inferior and medial temporal

cortex [135]. In conjunction with the finding that early-onset AD patients had more [$^{11}$C]-PBR28 binding than late-onset AD patients [137], these findings imply that microglia activation is a marker of neurodegeneration across various subtypes of AD. However, the variations in sample sizes and quantification methodologies make it difficult to convey a clear message.

### 14.6.2 Cognitive Deficits of AD

Neuroinflammation and cognition have also been linked by researchers. The majority of research investigated this association through using TSPO binding and global cognition measured by cognitive tests such as the mini-mental state examination (MMSE). In the case of correlations with cognitive deficits, the use of [$^{11}$C]-PBR28 binding revealed negative associations between TSPO binding and cognitive tests scores of MMSE, Clinical Dementia Rating Scale (CDR) Sum of Boxes, Trail Making Part B, Logical Memory Immediate, and Block Design [135]. Additional experiments utilizing [$^{11}$C]-DPA-713 or [$^{18}$F]-FEPPA have proposed that impairments in visuospatial function and linguistic ability were associated with TSPO binding in the parietal cortex and posterior limb of the internal capsule [78, 138]. Moreover, some studies demonstrated no correlation between TSPO binding and cognitive impairments [79, 83, 133], and subsequent research with a larger sample size demonstrated that [$^{18}$F]-DPA-714 binding was positively associated with MMSE score [93], which could imply a protective role of neuroinflammation in the early or even preclinical stages of AD.

Collectively, it is unknown whether neuroinflammatory alterations correspond with the degree of cognitive impairment. We may assume that there is a correlation between neuroinflammation and cognitive impairment; however, this link may not become evident until late in the illness. Additionally, cognitive deficits are probably impacted by several pathologies, such as the existence of neurofibrillary tangles.

### 14.6.3 Amyloid Deposition

Reactive microglia have been shown to colocalize closely with Aβ plaques in the brains of AD patients in postmortem studies, suggesting a possible connection between amyloid deposition and microglial activation [141, 142]. The amyloid cascade-inflammation theory suggested sequential development of amyloid plaques, microglial activation, and neurofibrillary [143]. Thus, given the discovery of activated microglial cells grouped around Aβ plaques, several studies investigated if the spatial connection can be measured in vivo using the most widely used amyloid radioligand, [$^{11}$C]-PIB.

The topic of a possible association between neuroinflammation and amyloid burden conducted by in vivo PET imaging is currently being debated. The first research to examine this link found no association between [$^{11}$C]-PK11195 and [$^{11}$C]-PIB in 13 AD patients and 14 healthy individuals [128]. Two subsequent

investigations using [$^{11}$C]-PK11195 and [$^{11}$C]-PIB in AD also revealed conflicting results; one group observed no spatial association [131], while the other group discovered a negative correlation in the posterior cingulate in AD patients [132]. The absence of geographical correlations suggested that microglial activation may be affected by other diseases such as tangle buildup. Additionally, they hypothesized that beta-amyloid oligomers might be involved in activated microglia, which would account for the absence of association, as [$^{11}$C]-PIB binds to fibrillar Aβ rather than oligomer Aβ. A more recent longitudinal study using second-generation TSPO ligands reported positive correlation between [$^{11}$C]-PIB and [$^{11}$C]-PBR28 in AD patients [139].

## 14.7 PET Imaging of Neuroinflammation in MCI

While most in vivo AD investigations have indicated an increase in TSPO binding, the precise timing of neuroinflammation throughout disease development is still unknown. To gain a better understanding, MCI, characterized as a stage between normal aging and dementia [144], was the subject of a neuroinflammation study to conduct an in vivo investigation of individuals who have not yet reached the advanced stage of cognitive impairment. The rate of advancement from MCI to dementia is 10–15% per year, which is substantially more significant than the rate of conversion in the general population, which is estimated to be 1–2% each year [145]. MCI is classified into two subtypes: amnestic (aMCI), which refers to memory impairments, and non-amnestic (naMCI), which refers to deficits in non-memory domains such as executive function, language, or visuospatial abilities [146]. Between the two kinds, aMCI is considered to represent the prodromal stage of AD, as persons with aMCI are more likely to develop to AD [147, 148].

In MCI, various TSPO radioligands have been utilized to measure neuroinflammation in vivo, similar to what has been done in AD research. Using the prototypical radioligand [$^{11}$C]-PK11195, conflicting findings have been published in the literature. Two studies found no significant difference in [$^{11}$C]-PK11195 binding between healthy controls and an MCI population [131, 133]. In contrast, according to the findings, one study of 14 individuals with aMCI discovered higher [$^{11}$C]-PK11195 uptake in 38% of the patients [129]. After multiple comparisons were corrected for, only the [$^{11}$C]-PK11195 binding in the frontal cortex remained substantially higher. In addition, increased binding was detected in the posterior cingulate, anterior cingulate, and frontal cortex.

Additionally, several studies employed second-generation radioligands for TSPO to investigate neuroinflammation in MCI [135, 149]. [$^{11}$C]-DAA1106 binding was shown to be significantly increased in the medial prefrontal cortex, cerebellum, parietal cortex, anterior cingulate cortex, lateral temporal cortex, and striatum, among other areas [149]. Individuals whose [$^{11}$C]-DAA1106 binding was more significant than the healthy control progressed to dementia within 5 years, which was a surprising finding. In addition, in later research comparing MCI-AD patients and demented-AD patients, neuroinflammation was shown to be stronger in

MCI-AD patients, supporting the notion that neuroinflammation is greater during the early stages of the illness [16]. Using [$^{11}$C]-PBR28, on the other hand, no changes were detected between MCI patients and healthy controls. The latter research was the first to account for the influence of the TSPO genotype on the outcome of the experiment (rs6971). Four HABs and six MABs were identified among the ten MCI patients who were included in the study [135].

Researchers studying neuroinflammation in MCI have also examined the possibility of seeing the geographical connection between microglia and amyloid plaques in vivo [129, 131, 135]. Two investigations using [$^{11}$C]-PIB and [$^{11}$C]-PK11195 in MCI and AD populations discovered no geographic associations between the two radioligands [129, 131]. In comparison, more recent research employing the second-generation radioligand [$^{11}$C]-PBR28 discovered a spatial connection between the radioligands' binding in the inferior parietal lobule, superior temporal cortex, precuneus, hippocampus, and parahippocampal gyrus. These associations, however, were established using partial volume adjusted data and by combining the two patient groups of AD and MCI [135]. Taking the previous five TSPO PET investigations in this group into account, it is evident that the presence of neuroinflammation during this prodromal period remains unknown.

Correlations between TSPO binding and cognitive impairment have also been investigated in research exploring neuroinflammation in MCI populations, with most studies finding negative results [129, 133, 149]. The only study involving both AD and MCI to establish a correlation between binding and cognition included patients with AD and MCI (19 AD and 10 MCI) and found a significant association between binding and cognitive impairment [89, 135].

## 14.8 Clinical Implications of Molecular Imaging for Neuroinflammation in AD and MCI

There are no effective interventions for treating the degenerative course of AD. Thus, early diagnosis of AD based on molecular imaging for neuroinflammation will be beneficial when effective treatments based on neuroinflammation become available [150]. Early clinical studies using nonsteroidal anti-inflammatory drugs (NSAIDs) in mild-moderate AD patients have shown considerable protection against progression of cognitive impairment over a 6-month period [151]. However, large-scale clinical research examining the preventive benefits of anti-inflammatories associated with progressive cognitive loss in mild to moderate AD has mixed results with poor side effects such as excess cardiovascular risk [152–157]. In addition, another study that explored whether anti-inflammatories were effective at delaying the onset of AD in high-risk groups showed that treated groups tended to have more impaired cognitive functions [158]. These results show that the use of anti-inflammatories without additional evaluation of neuroinflammation based on the conventional diagnostic system is not adequate.

Despite negative results, further studies investigating data from studies on the efficacy of anti-inflammatories in AD have provided implications for the use of

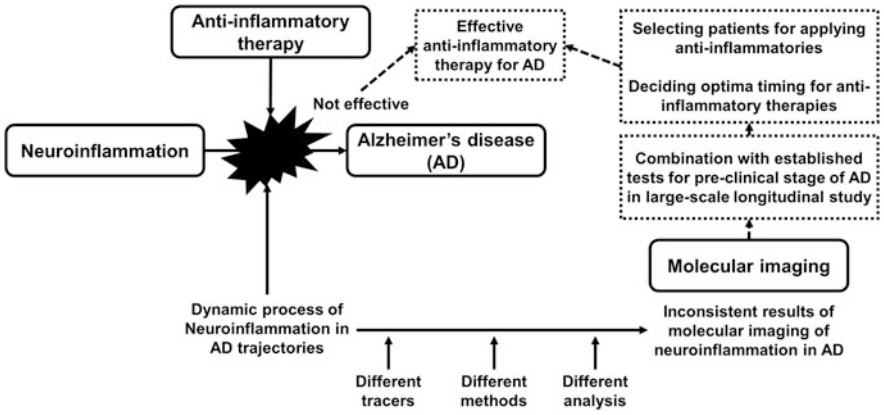


**Fig. 14.1** The clinical implications of molecular imaging for neuroinflammation in Alzheimer's disease, including the current status and future improvement directions. The contents included in the dotted line are the contents of future improvement directions. Ineffectiveness of anti-inflammatories in treating Alzheimer's disease to date and inconsistent results from molecular imaging appear to be due to dynamic process of neuroinflammation. By conducting a large-scale longitudinal study using molecular imaging including the early preclinical stage of Alzheimer's disease through established tests, it will be possible to obtain comprehensive information on the appropriate patient and the appropriate time to apply anti-inflammatories

anti-inflammatories. When investigating the data with subgroup analysis, anti-inflammatories were protective before symptom onset but harmful after symptom onset, and their effects varied according to the rate of decline [159, 160]. In animal models of AD, microglial activation was presented at the pre-plaque stage [161]. And, in another study, the injection of Aβ into the brain alone did not induce amyloid pathology, but it was reported that amyloid plaque formation was detected when lipopolysaccharide, which induces systemic inflammation, was administered alongside it [162]. Additionally, as discussed in the previous section, studies using molecular imaging studies in people with MCI reported increased microglial activation in the absence of amyloid tracer uptake [93, 137]. Therefore, it is necessary to investigate neuroinflammation focusing on early AD changes, and molecular imaging for neuroinflammation could play an important role in this process (Fig. 14.1).

To examine neuroinflammation in early AD, it seems investigating several factors at the same time will be needed. Several tests, such as tau in blood and CSF, PET scanning using [$^{11}$C]-PIB, and early genetic testing for the ApoE ε4 allele, can already be used to diagnose early stages of AD, so it is necessary to combine these with neuroinflammation molecular imaging. Epidemiological investigations demonstrated an association between NSAID usage and the ApoE ε4 genotype [163, 164], and a large cohort study demonstrated that carriers of ApoE ε4 with early AD progression benefited more from NSAIDs in terms of lowering the chance of developing AD [165]. Additionally, a clinical experiment that provided ibuprofen and esomeprazole to individuals with mild to moderate AD for 1 year discovered that participants with the ApoE ε4 allele experienced reduced cognitive deterioration

[166]. Molecular imaging will play an important role in providing comprehensive information on neuroinflammation in AD in view of the different modalities that can be performed simultaneously with multiple tests.

Although inconsistent results related to cognitive decline and subtypes concerning neuroinflammation in AD patients have been reported, a meta-analysis so far provided helpful insights for several studies introduced in this chapter. This study reported a detrimental role of microglial activation in the later stages of AD based on a significant correlation between neuroinflammation and cognitive decline, which are more prominent in AD [167]. However, since different tracers, the methods of tracer analysis, and other image analysis strategies all significantly influence the results of PET imaging studies [78, 136, 168], it is difficult to simplify the inconsistent results shown in several studies. Cohort studies involving both prodromal and dementia stages of AD show that neuroinflammation in AD is a dynamic process [150]. In particular, because microglial activation can also play a positive role to degrade and clear Aβ [169], the role of microglial activation in AD trajectories cannot be explained by a single process [150]. Results regarding distinct patterns of microglial activation in people with MCI or AD are thought to reflect this complexity [134]. It is thought that this complexity of neuroinflammation in AD trajectories contributes to the inconsistency of the molecular imaging results examined so far. Therefore, a large-scale longitudinal study using the same tracer and analysis method in both MCI and AD is needed to understand the dynamic role of neuroinflammation. These results may be helpful for deciding the optimal timing of anti-inflammatory therapies.

## 14.9 Conclusion

According to current research, neuroinflammation appears to play a critical role in the development and progression of AD. Molecular imaging is a helpful tool to identify such neuroinflammation, and PET imaging is the best-known molecular imaging. In PET imaging, the main targets to investigate in neuroinflammation are microglia and astrocytes, and various radioligands have been utilized. Studies examining neuroinflammation in AD and MCI showed significant differences from the normal group, and in addition, significant associations were found with clinically important subtypes, cognitive impairment, and amyloid deposits in AD. However, inconsistent results have been continuously reported. These results are thought to be due to the dynamic course of neuroinflammation in AD trajectories. Considering this complexity, to utilize anti-inflammatories prophylactically and therapeutically in AD, it will be necessary to longitudinally examine neuroinflammation in early-state AD together with various other markers.

## References


1. Mayeux R, Stern Y. Epidemiology of Alzheimer disease. Cold Spring Harb Perspect Med. 2012;2(8):a006239. https://pubmed.ncbi.nlm.nih.gov/22908189
2. Brookmeyer R, Johnson E, Ziegler-Graham K, Arrighi HM. Forecasting the global burden of Alzheimer's disease. Alzheimers Dement. 2007;3(3):186–91. http://www.sciencedirect.com/science/article/pii/S155252600700475X.
3. Ziegler-Graham K, Brookmeyer R, Johnson E, Arrighi HM. Worldwide variation in the doubling time of Alzheimer's disease incidence rates. Alzheimers Dement. 2008;4(5):316–23. https://doi.org/10.1016/j.jalz.2008.05.2479.
4. Mebane-Sims I. Alzheimer's Association, 2018 Alzheimer's disease facts and figures. Alzheimers Dement. 2018;14(3):367–429.
5. Kawas C, Gray S, Brookmeyer R, Fozard J, Zonderman A. Age-specific incidence rates of Alzheimer's disease: the Baltimore longitudinal study of aging. Neurology. 2000;54(11):2072–7.
6. Borenstein AR. Survival and mortality in Alzheimer's disease. In: Borenstein AR, editor. Alzheimer's disease: life course perspectives on risk reduction. Amsterdam: Elsevier; 2016. p. 89–94.
7. Jia J, Wei C, Chen S, Li F, Tang Y, Qin W, et al. The cost of Alzheimer's disease in China and re-estimation of costs worldwide. Alzheimer's Dement. 2018;14:483–91.
8. Atri A. The Alzheimer's disease clinical spectrum: diagnosis and management. Med Clin North Am. 2019;103(2):263–93. https://doi.org/10.1016/j.mcna.2018.10.009.
9. Association AP. Diagnostic and statistical manual of mental disorders (DSM-5®). Washington, DC: American Psychiatric Pub; 2013.
10. Braak H, Braak E. Neuropathological stageing of Alzheimer-related changes. Acta Neuropathol. 1991;82:239–59.
11. Hardy J, Selkoe DJ. The amyloid hypothesis of Alzheim progress and problems on the road to. Science. 2002;297(5580):353–6.
12. Herrup K. The case for rejecting the amyloid cascade hypothesis. Nat Neurosci. 2015;18(6):794–9.
13. Kinney JW, Bemiller SM, Murtishaw AS, Leisgang AM, Salazar AM, Lamb BT. Inflammation as a central mechanism in Alzheimer's disease. Alzheimer's Dement Transl Res Clin Interv. 2018;4:575–90. http://www.sciencedirect.com/science/article/pii/S2352873718300490.
14. Schain M, Kreisl WC. Neuroinflammation in neurodegenerative disorders—a review. Curr Neurol Neurosci Rep. 2017;17(3):25. https://doi.org/10.1007/s11910-017-0733-2.
15. Kreisl WC, Kim M-J, Coughlin JM, Henter ID, Owen DR, Innis RB. PET imaging of neuroinflammation in neurological disorders. Lancet Neurol. 2020;19(11):940–50. https://linkinghub.elsevier.com/retrieve/pii/S147444222030346X.
16. Lagarde J, Sarazin M, Bottlaender M. In vivo PET imaging of neuroinflammation in Alzheimer's disease. J Neural Transm. 2018;125(5):847–67. http://link.springer.com/10.1007/s00702-017-1731-x.
17. Schwartz M, Deczkowska A. Neurological disease as a failure of brain–immune crosstalk: the multiple faces of neuroinflammation. Trends Immunol. 2016;37(10):668–79. https://doi.org/10.1016/j.it.2016.08.001.
18. Prokop S, Miller KR, Heppner FL. Microglia actions in Alzheimer's disease. Acta Neuropathol. 2013;126(4):461–77.
19. Schwab C, McGeer PL. Inflammatory aspects of Alzheimer disease and other neurodegenerative disorders. J Alzheimers Dis. 2008;13:359–69.
20. Gomez-Nicola D, Boche D. Post-mortem analysis of neuroinflammatory changes in human Alzheimer's disease. Alzheimers Res Ther. 2015;7(1):42. https://doi.org/10.1186/s13195-015-0126-1.

21. Johnston H, Boutin H, Allan SM. Assessing the contribution of inflammation in models of Alzheimer's disease. Biochem Soc Trans. 2011;39(4):886–90. https://doi.org/10.1042/BST0390886.
22. Heneka MT, Carson MJ, El Khoury J, Landreth GE, Brosseron F, Feinstein DL, et al. Neuroinflammation in Alzheimer's disease. Lancet Neurol. 2015;14(4):388–405. http://www.sciencedirect.com/science/article/pii/S1474442215700165.
23. Ries M, Sastre M. Mechanisms of Aβ clearance and degradation by glial cells. Front Aging Neurosci. 2016;8:1–9.
24. Mandrekar S, Jiang Q, Lee CYD, Koenigsknecht-Talboo J, Holtzman DM, Landreth GE. Microglia mediate the clearance of soluble aβ through fluid phase macropinocytosis. J Neurosci. 2009;29(13):4252–62.
25. Phelps ME. Positron emission tomography provides molecular imaging of biological processes. Proc Natl Acad Sci. 2000;97(16):9226–33. http://www.pnas.org/content/97/16/9226.abstract.
26. Owen DRJ, Matthews PM. Chapter 2 - Imaging brain microglial activation using positron emission tomography and translocator protein-specific radioligands. In: Guest PC, Bahn S, editors. Biomarkers of neurological and psychiatric disease. London: Academic Press; 2011. p. 19–39. https://www.sciencedirect.com/science/article/pii/B978012387718500002X.
27. Colonna M, Butovsky O. Microglia function in the central nervous system during health and neurodegeneration. Annu Rev Immunol. 2017;35:441–68.
28. Paolicelli RC, Gross CT. Microglia in development: linking brain wiring to brain environment. Neuron Glia Biol. 2011;7(1):77–83.
29. Venneti S, Wiley CA, Kofler J. Imaging microglial activation during neuroinflammation and Alzheimer's disease. J NeuroImmune Pharmacol. 2009;4(2):227–43.
30. Keren-Shaul H, Spinrad A, Weiner A, Matcovitch-Natan O, Dvir-Szternfeld R, Ulland TK, et al. A unique microglia type associated with restricting development of Alzheimer's disease. Cell. 2017;169(7):1276–1290.e17.
31. Boche D, Gerhard A, Rodriguez-Vieitez E, Faculty on behalf of the M. Prospects and challenges of imaging neuroinflammation beyond TSPO in Alzheimer's disease. Eur J Nucl Med Mol Imaging. 2019;46(13):2831–47. https://doi.org/10.1007/s00259-019-04462-w.
32. Felsky D, Roostaei T, Nho K, Risacher SL, Bradshaw EM, Petyuk V, et al. Neuropathological correlates and genetic architecture of microglial activation in elderly human brain. Nat Commun. 2019;10(1):409.
33. Sofroniew MV, Vinters HV. Astrocytes: biology and pathology. Acta Neuropathol. 2010;119(1):7–35.
34. Attwell D, Buchan AM, Charpak S, Lauritzen M, Macvicar BA, Newman EA. Glial and neuronal control of brain blood flow. Nature. 2010;468(7321):232–43.
35. Pekny M, Pekna M, Messing A, Steinhäuser C, Lee J-M, Parpura V, et al. Astrocytes: a central element in neurological diseases. Acta Neuropathol. 2016;131(3):323–45.
36. Eroglu C, Barres BA. Regulation of synaptic connectivity by glia. Nature. 2010;468(7321):223–31.
37. Jessen NA, Munk ASF, Lundgaard I, Nedergaard M. The glymphatic system: a beginner's guide. Neurochem Res. 2015;40(12):2583–99.
38. Tarasoff-Conway JM, Carare RO, Osorio RS, Glodzik L, Butler T, Fieremans E, et al. Clearance systems in the brain-implications for Alzheimer disease. Nat Rev Neurol. 2015;11(8):457–70.
39. Sofroniew MV. Multiple roles for astrocytes as effectors of cytokines and inflammatory mediators. Neuroscientist. 2014;20(2):160–72.
40. Thal DR. The role of astrocytes in amyloid β-protein toxicity and clearance. Exp Neurol. 2012;236(1):1–5.
41. Wyss-Coray T, Loike JD, Brionne TC, Lu E, Anankov R, Yan F, et al. Adult mouse astrocytes degrade amyloid-β in vitro and in situ. Nat Med. 2003;9(4):453–7. https://doi.org/10.1038/nm838.

42. Allaman I, Gavillet M, Bélanger M, Laroche T, Viertl D, Lashuel HA, et al. Amyloid-beta aggregates cause alterations of astrocytic metabolic phenotype: impact on neuronal viability. J Neurosci. 2010;30(9):3326–38.
43. Song W, Zhou LJ, Zheng SX, Zhu XZ. Amyloid-beta 25-35 peptide induces expression of monoamine oxidase B in cultured rat astrocytes. Acta Pharmacol Sin. 2000;21(6):557–63.
44. Jo S, Yarishkin O, Hwang YJ, Chun YE, Park M, Woo DH, et al. GABA from reactive astrocytes impairs memory in mouse models of Alzheimer's disease. Nat Med. 2014;20(8):886–96.
45. Funato H, Yoshimura M, Yamazaki T, Saido TC, Ito Y, Yokofujita J, et al. Astrocytes containing amyloid beta-protein (Abeta)-positive granules are associated with Abeta40-positive diffuse plaques in the aged human brain. Am J Pathol. 1998;152(4):983–92.
46. Thal DR, Schultz C, Dehghani F, Yamaguchi H, Braak H, Braak E. Amyloid beta-protein (Abeta)-containing astrocytes are located preferentially near N-terminal-truncated Abeta deposits in the human entorhinal cortex. Acta Neuropathol. 2000;100(6):608–17.
47. Wyss-Coray T, Loike JD, Brionne TC, Lu E, Anankov R, Yan F, et al. Adult mouse astrocytes degrade amyloid-beta in vitro and in situ. Nat Med. 2003;9(4):453–7.
48. Schöll M, Carter SF, Westman E, Rodriguez-Vieitez E, Almkvist O, Thordardottir S, et al. Early astrocytosis in autosomal dominant Alzheimer's disease measured in vivo by multi-tracer positron emission tomography. Sci Rep. 2015;5:16404.
49. Perry VH, Nicoll JAR, Holmes C. Microglia in neurodegenerative disease. Nat Rev Neurol. 2010;6(4):193–201. https://doi.org/10.1038/nrneurol.2010.17.
50. Braestrup C, Squires RF. Specific benzodiazepine receptors in rat brain characterized by high-affinity (3H)diazepam binding. Proc Natl Acad Sci. 1977;74(9):3805–9. http://www.pnas.org/content/74/9/3805.abstract.
51. McEnery MW, Snowman AM, Trifiletti RR, Snyder SH. Isolation of the mitochondrial benzodiazepine receptor: association with the voltage-dependent anion channel and the adenine nucleotide carrier. Proc Natl Acad Sci U S A. 1992;89(8):3170–4.
52. Casellas P, Galiegue S, Basile AS. Peripheral benzodiazepine receptors and mitochondrial function. Neurochem Int. 2002;40(6):475–86.
53. Venneti S, Lopresti BJ, Wiley CA. The peripheral benzodiazepine receptor (Translocator protein 18kDa) in microglia: from pathology to imaging. Prog Neurobiol. 2006;80(6):308–22.
54. Liu G-J, Middleton RJ, Hatty CR, Kam WW-Y, Chan R, Pham T, et al. The 18 kDa translocator protein, microglia and neuroinflammation. Brain Pathol. 2014;24(6):631–53.
55. Papadopoulos V, Baraldi M, Guilarte TR, Knudsen TB, Lacapère J-J, Lindemann P, et al. Translocator protein (18kDa): new nomenclature for the peripheral-type benzodiazepine receptor based on its structure and molecular function. Trends Pharmacol Sci. 2006;27(8):402–9. http://www.sciencedirect.com/science/article/pii/S0165614706001532.
56. Chen M-K, Guilarte TR. Translocator protein 18 kDa (TSPO): molecular sensor of brain injury and repair. Pharmacol Ther. 2008;118(1):1–17. https://www.sciencedirect.com/science/article/pii/S0163725808000168.
57. Costa B, Da Pozzo E, Martini C. 18-kDa translocator protein association complexes in the brain: from structure to function. Biochem Pharmacol. 2020;177:114015. https://www.sciencedirect.com/science/article/pii/S0006295220302434.
58. Nutma E, Ceyzériat K, Amor S, Tsartsalis S, Millet P, Owen DR, et al. Cellular sources of TSPO expression in healthy and diseased brain. Eur J Nucl Med Mol Imaging. 2021;49:146. https://doi.org/10.1007/s00259-020-05166-2.
59. Gulyás B, Makkai B, Kása P, Gulya K, Bakota L, Várszegi S, et al. A comparative autoradiography study in post mortem whole hemisphere human brain slices taken from Alzheimer patients and age-matched controls using two radiolabelled DAA1106 analogues with high affinity to the peripheral benzodiazepine receptor (PBR) syst. Neurochem Int. 2009;54(1):28–36. https://www.sciencedirect.com/science/article/pii/S0197018608001629.

60. Weissman BA, Bolger GT, Isaac L, Paul SM, Skolnick P. Characterization of the binding of [3H]Ro 5-4864, a convulsant benzodiazepine, to guinea pig brain. J Neurochem. 1984;42(4):969–75.
61. Gavish M, Bachman I, Shoukrun R, Katz Y, Veenman L, Weisinger G, et al. Enigma of the peripheral benzodiazepine receptor. Pharmacol Rev. 1999;51(4):629–50.
62. Stephenson DT, Schober DA, Smalstig EB, Mincy RE, Gehlert DR, Clemens JA. Peripheral benzodiazepine receptors are colocalized with activated microglia following transient global forebrain ischemia in the rat. J Neurosci. 1995;15(7 Pt 2):5263–74.
63. Banati RB. Visualising microglial activation in vivo. Glia. 2002;40(2):206–17.
64. Kuhlmann AC, Guilarte TR. Cellular and subcellular localization of peripheral benzodiazepine receptors after trimethyltin neurotoxicity. J Neurochem. 2000;74(4):1694–704.
65. Venneti S, Wang G, Nguyen J, Wiley CA. The positron emission tomography ligand DAA1106 binds with high affinity to activated microglia in human neurological disorders. J Neuropathol Exp Neurol. 2008;67(10):1001–10.
66. Cosenza-Nashat M, Zhao M-L, Suh H-S, Morgan J, Natividad R, Morgello S, et al. Expression of the translocator protein of 18 kDa by microglia, macrophages and astrocytes based on immunohistochemical localization in abnormal human brain. Neuropathol Appl Neurobiol. 2009;35(3):306–28. https://doi.org/10.1111/j.1365-2990.2008.01006.x.
67. Lavisse S, Guillermier M, Hérard A-S, Petit F, Delahaye M, Van Camp N, et al. Reactive astrocytes overexpress TSPO and are detected by TSPO positron emission tomography imaging. J Neurosci. 2012;32(32):10809–18.
68. Vivash L, O'Brien TJ. Imaging microglial activation with TSPO PET: lighting up neurologic diseases? J Nucl Med. 2016;57(2):165–8.
69. Cagnin A, Brooks DJ, Kennedy AM, Gunn RN, Myers R, Turkheimer FE, et al. In-vivo measurement of activated microglia in dementia. Lancet. 2001;358(9280):461–7. http://www.sciencedirect.com/science/article/pii/S0140673601056252.
70. Pavese N, Gerhard A, Tai YF, Ho AK, Turkheimer F, Barker RA, et al. Microglial activation correlates with severity in Huntington disease: a clinical and PET study. Neurology. 2006;66(11):1638–43.
71. Politis M, Giannetti P, Su P, Turkheimer F, Keihaninejad S, Wu K, et al. Increased PK11195 PET binding in the cortex of patients with MS correlates with disability. Neurology. 2012;79(6):523–30.
72. Gerhard A, Neumaier B, Elitok E, Glatting G, Ries V, Tomczak R, et al. In vivo imaging of activated microglia using [11 C] PK11195 and positron emission tomography in patients after ischemic stroke. Neuroreport. 2000;11(13):2957–60.
73. Ching ASC, Kuhnast B, Damont A, Roeda D, Tavitian B, Dollé F. Current paradigm of the 18-kDa translocator protein (TSPO) as a molecular target for PET imaging in neuroinflammation and neurodegenerative diseases. Insights Imaging. 2012;3(1):111–9.
74. Lockhart A, Davis B, Matthews JC, Rahmoune H, Hong G, Gee A, et al. The peripheral benzodiazepine receptor ligand PK11195 binds with high affinity to the acute phase reactant alpha1-acid glycoprotein: implications for the use of the ligand as a CNS inflammatory marker. Nucl Med Biol. 2003;30(2):199–206.
75. Fujita M, Imaizumi M, Zoghbi SS, Fujimura Y, Farris AG, Suhara T, et al. Kinetic analysis in healthy humans of a novel positron emission tomography radioligand to image the peripheral benzodiazepine receptor, a potential biomarker for inflammation. NeuroImage. 2008;40(1):43–52. https://www.sciencedirect.com/science/article/pii/S1053811907010488.
76. Kreisl WC, Fujita M, Fujimura Y, Kimura N, Jenko KJ, Kannan P, et al. Comparison of [11C]-(R)-PK 11195 and [11C]PBR28, two radioligands for translocator protein (18 kDa) in human and monkey: implications for positron emission tomographic imaging of this inflammation biomarker. NeuroImage. 2010;49(4):2924–32. https://www.sciencedirect.com/science/article/pii/S1053811909012427.
77. Chauveau F, Van Camp N, Dollé F, Kuhnast B, Hinnen F, Damont A, et al. Comparative evaluation of the translocator protein radioligands 11C-DPA-713, 18F-DPA-714, and

11C-PK11195 in a rat model of acute neuroinflammation. J Nucl Med. 2009;50(3):468–76. http://jnm.snmjournals.org/cgi/content/short/50/3/468. Accessed 15 Mar 2020.
78. Yokokura M, Terada T, Bunai T, Nakaizumi K, Takebayashi K, Iwata Y, et al. Depiction of microglial activation in aging and dementia: positron emission tomography with [(11)C] DPA713 versus [(11)C](R)PK11195. J Cereb Blood Flow Metab. 2017;37(3):877–89.
79. Yasuno F, Ota M, Kosaka J, Ito H, Higuchi M, Doronbekov TK, et al. Increased binding of peripheral benzodiazepine receptor in Alzheimer's disease measured by positron emission tomography with [11C]DAA1106. Biol Psychiatry. 2008;64(10):835–41. http://www.sciencedirect.com/science/article/pii/S000632230800499X.
80. Chauveau F, Boutin H, Van Camp N, Dollé F, Tavitian B. Nuclear imaging of neuroinflammation: a comprehensive review of [11C]PK11195 challengers. Eur J Nucl Med Mol Imaging. 2008;35(12):2304–19. https://doi.org/10.1007/s00259-008-0908-9.
81. Fujimura Y, Ikoma Y, Yasuno F, Suhara T, Ota M, Matsumoto R, et al. Quantitative analyses of 18F-FEDAA1106 binding to peripheral benzodiazepine receptors in living human brain. J Nucl Med. 2006;47(1):43–50.
82. Varrone A, Mattsson P, Forsberg A, Takano A, Nag S, Gulyás B, et al. In vivo imaging of the 18-kDa translocator protein (TSPO) with [18F]FEDAA1106 and PET does not show increased binding in Alzheimer's disease patients. Eur J Nucl Med Mol Imaging. 2013;40(6):921–31. https://doi.org/10.1007/s00259-013-2359-1.
83. Varrone A, Oikonen V, Forsberg A, Joutsa J, Takano A, Solin O, et al. Positron emission tomography imaging of the 18-kDa translocator protein (TSPO) with [18F]FEMPA in Alzheimer's disease patients and control subjects. Eur J Nucl Med Mol Imaging. 2015;42(3):438–46. https://doi.org/10.1007/s00259-014-2955-8.
84. Rusjan PM, Wilson AA, Bloomfield PM, Vitcu I, Meyer JH, Houle S, et al. Quantitation of translocator protein binding in human brain with the novel radioligand [18F]-FEPPA and positron emission tomography. J Cereb Blood Flow Metab. 2011;31(8):1807–16.
85. Gulyás B, Tóth M, Schain M, Airaksinen A, Vas Á, Kostulas K, et al. Evolution of microglial activation in ischaemic core and peri-infarct regions after stroke: a PET study with the TSPO molecular imaging biomarker [$^{11}$C]vinpocetine. J Neurol Sci. 2012;320(1):110–7. https://doi.org/10.1016/j.jns.2012.06.026.
86. Gulyás B, Vas A, Tóth M, Takano A, Varrone A, Cselényi Z, et al. Age and disease related changes in the translocator protein (TSPO) system in the human brain: positron emission tomography measurements with [11C]vinpocetine. NeuroImage. 2011;56(3):1111–21.
87. Owen DR, Yeo AJ, Gunn RN, Song K, Wadsworth G, Lewis A, et al. An 18-kDa translocator protein (TSPO) polymorphism explains differences in binding affinity of the PET radioligand PBR28. J Cereb Blood Flow Metab. 2012;32(1):1–5.
88. Guo Q, Colasanti A, Owen DR, Onega M, Kamalakaran A, Bennacef I, et al. Quantification of the specific translocator protein signal of 18F-PBR111 in healthy humans: a genetic polymorphism effect on in vivo binding. J Nucl Med. 2013;54(11):1915–23.
89. Kreisl WC, Jenko KJ, Hines CS, Lyoo CH, Corona W, Morse CL, et al. A genetic polymorphism for translocator protein 18 kDa affects both in vitro and in vivo radioligand binding in human brain to this putative biomarker of neuroinflammation. J Cereb Blood Flow Metab. 2013;33(1):53–8.
90. Owen DR, Howell OW, Tang S-P, Wells LA, Bennacef I, Bergstrom M, et al. Two binding sites for [3H]PBR28 in human brain: implications for TSPO PET imaging of neuroinflammation. J Cereb Blood Flow Metab. 2010;30(9):1608–18.
91. Knezevic D, Mizrahi R. Molecular imaging of neuroinflammation in Alzheimer's disease and mild cognitive impairment. Prog Neuro-Psychopharmacol Biol Psychiatry. 2018;80:123–31. https://doi.org/10.1016/j.pnpbp.2017.05.007.
92. Fan Z, Harold D, Pasqualetti G, Williams J, Brooks DJ, Edison P. Can Studies of neuroinflammation in a TSPO genetic subgroup (HAB or MAB) be applied to the entire AD cohort? J Nucl Med. 2015;56(5):707–13.

93. Hamelin L, Lagarde J, Dorothée G, Leroy C, Labit M, Comley RA, et al. Early and protective microglial activation in Alzheimer's disease: a prospective study using 18 F-DPA-714 PET imaging. Brain. 2016;139(4):1252–64. https://doi.org/10.1093/brain/aww017.
94. Srinivasan K, Friedman BA, Larson JL, Lauffer BE, Goldstein LD, Appling LL, et al. Untangling the brain's neuroinflammatory and neurodegenerative transcriptional responses. Nat Commun. 2016;7:11295.
95. Amenta PS, Jallo JI, Tuma RF, Hooper DC, Elliott MB. Cannabinoid receptor type-2 stimulation, blockade, and deletion alter the vascular inflammatory responses to traumatic brain injury. J Neuroinflammation. 2014;11(1):191. https://doi.org/10.1186/s12974-014-0191-6.
96. Ahmad R, Postnov A, Bormans G, Versijpt J, Vandenbulcke M, Van Laere K. Decreased in vivo availability of the cannabinoid type 2 receptor in Alzheimer's disease. Eur J Nucl Med Mol Imaging. 2016;43(12):2219–27.
97. Slavik R, Müller Herde A, Haider A, Krämer SD, Weber M, Schibli R, et al. Discovery of a fluorinated 4-oxo-quinoline derivative as a potential positron emission tomography radiotracer for imaging cannabinoid receptor type 2. J Neurochem. 2016;138(6):874–86.
98. Shukuri M, Takashima-Hirano M, Tokuda K, Takashima T, Matsumura K, Inoue O, et al. In vivo expression of cyclooxygenase-1 in activated microglia and macrophages during neuroinflammation visualized by PET with 11C- ketoprofen methyl ester. J Nucl Med. 2011;52(7):1094–101.
99. Bannwarth B, Netter P, Pourel J, Royer RJ, Gaucher A. Clinical pharmacokinetics of nonsteroidal anti-inflammatory drugs in the cerebrospinal fluid. Biomed Pharmacother. 1989;43(2):121–6. https://www.sciencedirect.com/science/article/pii/0753332289901406.
100. Ohnishi A, Senda M, Yamane T, Sasaki M, Mikami T, Nishio T, et al. Human whole-body biodistribution and dosimetry of a new PET tracer, [(11)C]ketoprofen methyl ester, for imagings of neuroinflammation. Nucl Med Biol. 2014;41(7):594–9.
101. Ohnishi A, Senda M, Yamane T, Mikami T, Nishida H, Nishio T, et al. Exploratory human PET study of the effectiveness of (11)C-ketoprofen methyl ester, a potential biomarker of neuroinflammatory processes in Alzheimer's disease. Nucl Med Biol. 2016;43(7):438–44.
102. Albrecht DS, Granziera C, Hooker JM, Loggia ML. In vivo imaging of human neuroinflammation. ACS Chem Neurosci. 2016;7(4):470–83.
103. Wang HY, Lee DH, D'Andrea MR, Peterson PA, Shank RP, Reitz AB. beta-Amyloid(1-42) binds to alpha7 nicotinic acetylcholine receptor with high affinity. Implications for Alzheimer's disease pathology. J Biol Chem. 2000;275(8):5626–32.
104. Kalkman HO, Feuerbach D. Modulatory effects of α7 nAChRs on the immune system and its relevance for CNS disorders. Cell Mol Life Sci. 2016;73(13):2511–30.
105. Hillmer AT, Li S, Zheng M-Q, Scheunemann M, Lin S-F, Nabulsi N, et al. PET imaging of α(7) nicotinic acetylcholine receptors: a comparative study of [(18)F]ASEM and [(18)F]DBT-10 in nonhuman primates, and further evaluation of [(18)F]ASEM in humans. Eur J Nucl Med Mol Imaging. 2017;44(6):1042–50.
106. Gourine AV, Kasparov S. Astrocytes as brain interoceptors. Exp Physiol. 2011;96(4):411–6.
107. De Strooper B, Karran E. The cellular phase of Alzheimer's disease. Cell. 2016;164(4):603–15.
108. Hirvonen J, Kailajärvi M, Haltia T, Koskimies S, Någren K, Virsu P, et al. Assessment of MAO-B occupancy in the brain with PET and [11C]-L-deprenyl-D2: a dose-finding study with a novel MAO-B inhibitor, EVT 301. Clin Pharmacol Ther. 2009;85(5):506–12.
109. Sturm S, Forsberg A, Nave S, Stenkrona P, Seneca N, Varrone A, et al. Positron emission tomography measurement of brain MAO-B inhibition in patients with Alzheimer's disease and elderly controls after oral administration of sembragiline. Eur J Nucl Med Mol Imaging. 2017;44(3):382–91.
110. Gulyás B, Pavlova E, Kása P, Gulya K, Bakota L, Várszegi S, et al. Activated MAO-B in the brain of Alzheimer patients, demonstrated by [11C]-L-deprenyl using whole hemisphere autoradiography. Neurochem Int. 2011;58(1):60–8.

111. Tong J, Rathitharan G, Meyer JH, Furukawa Y, Ang L-C, Boileau I, et al. Brain monoamine oxidase B and A in human parkinsonian dopamine deficiency disorders. Brain. 2017;140(9):2460–74.
112. Rodriguez-Vieitez E, Carter SF, Chiotis K, Saint-Aubert L, Leuzy A, Schöll M, et al. Comparison of early-phase 11C-deuterium-l-deprenyl and 11C-pittsburgh compound B PET for assessing brain perfusion in Alzheimer disease. J Nucl Med. 2016;57(7):1071–7.
113. Santillo AF, Gambini JP, Lannfelt L, Långström B, Ulla-Marja L, Kilander L, et al. In vivo imaging of astrocytosis in Alzheimer's disease: an $^{11}$C-L-deuteriodeprenyl and PIB PET study. Eur J Nucl Med Mol Imaging. 2011;38(12):2202–8.
114. Carter SF, Schöll M, Almkvist O, Wall A, Engler H, Långström B, et al. Evidence for astrocytosis in prodromal Alzheimer disease provided by 11C-deuterium-L-deprenyl: a multitracer PET paradigm combining 11C-pittsburgh compound B and 18F-FDG. J Nucl Med. 2012;53(1):37–46. http://jnm.snmjournals.org/content/53/1/37.abstract.
115. Rodriguez-Vieitez E, Saint-Aubert L, Carter SF, Almkvist O, Farid K, Schöll M, et al. Diverging longitudinal changes in astrocytosis and amyloid PET in autosomal dominant Alzheimer's disease. Brain. 2016;139(3):922–36.
116. Zimmer ER, Parent MJ, Souza DG, Leuzy A, Lecrux C, Kim H-I, et al. [(18)F]FDG PET signal is driven by astroglial glutamate transport. Nat Neurosci. 2017;20(3):393–5.
117. Magistretti PJ, Pellerin L. The contribution of astrocytes to the 18F-2-deoxyglucose signal in PET activation studies. Mol Psychiatry. 1996;1(6):445–52.
118. Pellerin L, Pellegri G, Bittar PG, Charnay Y, Bouras C, Martin JL, et al. Evidence supporting the existence of an activity-dependent astrocyte-neuron lactate shuttle. Dev Neurosci. 1998;20(4–5):291–9.
119. Carter SF, Chiotis K, Nordberg A, Rodriguez-Vieitez E. Longitudinal association between astrocyte function and glucose metabolism in autosomal dominant Alzheimer's disease. Eur J Nucl Med Mol Imaging. 2019;46(2):348–56.
120. Acosta C, Anderson HD, Anderson CM. Astrocyte dysfunction in Alzheimer disease. J Neurosci Res. 2017;95(12):2430–47.
121. Hefendehl JK, LeDue J, Ko RWY, Mahler J, Murphy TH, MacVicar BA. Mapping synaptic glutamate transporter dysfunction in vivo to regions surrounding Aβ plaques by iGluSnFR two-photon imaging. Nat Commun. 2016;7:13441.
122. Olabarria M, Noristani HN, Verkhratsky A, Rodríguez JJ. Age-dependent decrease in glutamine synthetase expression in the hippocampal astroglia of the triple transgenic Alzheimer's disease mouse model: mechanism for deficient glutamatergic transmission? Mol Neurodegener. 2011;6:55.
123. Pascual JM, Van Heertum RL, Wang D, Engelstad K, De Vivo DC. Imaging the metabolic footprint of Glut1 deficiency on the brain. Ann Neurol. 2002;52(4):458–64.
124. Vlassenko AG, Gordon BA, Goyal MS, Su Y, Blazey TM, Durbin TJ, et al. Aerobic glycolysis and tau deposition in preclinical Alzheimer's disease. Neurobiol Aging. 2018;67:95–8.
125. Mishina M, Ishiwata K, Naganawa M, Kimura Y, Kitamura S, Suzuki M, et al. Adenosine A (2A) receptors measured with [C]TMSX PET in the striata of Parkinson's disease patients. PLoS One. 2011;6(2):e17338.
126. Rissanen E, Virta JR, Paavilainen T, Tuisku J, Helin S, Luoto P, et al. Adenosine A2A receptors in secondary progressive multiple sclerosis: a [(11)C]TMSX brain PET study. J Cereb Blood Flow Metab. 2013;33(9):1394–401.
127. Versijpt JJ, Dumont F, Van Laere KJ, Decoo D, Santens P, Audenaert K, et al. Assessment of neuroinflammation and microglial activation in Alzheimer's disease with radiolabelled PK11195 and single photon emission computed tomography. A pilot study. Eur Neurol. 2003;50(1):39–47.
128. Edison P, Archer HA, Gerhard A, Hinz R, Pavese N, Turkheimer FE, et al. Microglia, amyloid, and cognition in Alzheimer's disease: an [11C](R)PK11195-PET and [11C]PIB-PET study. Neurobiol Dis. 2008;32(3):412–9. http://www.sciencedirect.com/science/article/pii/S0969996108001885.

129. Okello A, Edison P, Archer HA, Turkheimer FE, Kennedy J, Bullock R, et al. Microglial activation and amyloid deposition in mild cognitive impairment: a PET study. Neurology. 2009;72(1):56–62. https://pubmed.ncbi.nlm.nih.gov/19122031.
130. Groom GN, Junck L, Foster NL, Frey KA, Kuhl DE. PET of peripheral benzodiazepine binding sites in the microgliosis of Alzheimer's disease. J Nucl Med. 1995;36(12):2207–10.
131. Wiley CA, Lopresti BJ, Venneti S, Price J, Klunk WE, DeKosky ST, et al. Carbon 11-labeled pittsburgh compound B and carbon 11-labeled (R)-PK11195 positron emission tomographic imaging in Alzheimer disease. Arch Neurol. 2009;66(1):60–7.
132. Yokokura M, Mori N, Yagi S, Yoshikawa E, Kikuchi M, Yoshihara Y, et al. In vivo changes in microglial activation and amyloid deposits in brain regions with hypometabolism in Alzheimer's disease. Eur J Nucl Med Mol Imaging. 2011;38(2):343–51. https://doi.org/10.1007/s00259-010-1612-0.
133. Schuitemaker A, Kropholler MA, Boellaard R, van der Flier WM, Kloet RW, van der Doef TF, et al. Microglial activation in Alzheimer's disease: an (R)-[11C]PK11195 positron emission tomography study. Neurobiol Aging. 2013;34(1):128–36. http://www.sciencedirect.com/science/article/pii/S0197458012002722.
134. Fan Z, Brooks DJ, Okello A, Edison P. An early and late peak in microglial activation in Alzheimer's disease trajectory. Brain. 2017;140(3):792–803.
135. Kreisl WC, Lyoo CH, McGwier M, Snow J, Jenko KJ, Kimura N, et al. In vivo radioligand binding to translocator protein correlates with severity of Alzheimer's disease. Brain. 2013;136(7):2228–38. https://doi.org/10.1093/brain/awt145.
136. Lyoo CH, Ikawa M, Liow J-S, Zoghbi SS, Morse CL, Pike VW, et al. Cerebellum can serve as a pseudo-reference region in Alzheimer disease to detect neuroinflammation measured with PET radioligand binding to translocator protein. J Nucl Med. 2015;56(5):701–6. http://www.ncbi.nlm.nih.gov/pubmed/25766898. Accessed 15 Mar 2020.
137. Kreisl WC, Lyoo CH, Liow J-S, Wei M, Snow J, Page E, et al. (11)C-PBR28 binding to translocator protein increases with progression of Alzheimer's disease. Neurobiol Aging. 2016;44:53–61. https://pubmed.ncbi.nlm.nih.gov/27318133.
138. Suridjan I, Pollock BG, Verhoeff NPLG, Voineskos AN, Chow T, Rusjan PM, et al. In-vivo imaging of grey and white matter neuroinflammation in Alzheimer's disease: a positron emission tomography study with a novel radioligand, [18F]-FEPPA. Mol Psychiatry. 2015;20(12):1579–87. https://doi.org/10.1038/mp.2015.1.
139. Kreisl WC, Lyoo CH, Liow J-S, Snow J, Page E, Jenko KJ, et al. Distinct patterns of increased translocator protein in posterior cortical atrophy and amnestic Alzheimer's disease. Neurobiol Aging. 2017;51:132–40. https://www.sciencedirect.com/science/article/pii/S0197458016303128.
140. Cerami C, Crespi C, Della Rosa PA, Dodich A, Marcone A, Magnani G, et al. Brain changes within the visuo-spatial attentional network in posterior cortical atrophy. J Alzheimers Dis. 2015;43:385–95.
141. McGeer PL, Itagaki S, Tago H, McGeer EG. Reactive microglia in patients with senile dementia of the Alzheimer type are positive for the histocompatibility glycoprotein HLA-DR. Neurosci Lett. 1987;79(1–2):195–200.
142. Tooyama I, Kimura H, Akiyama H, McGeer PL. Reactive microglia express class I and class II major histocompatibility complex antigens in Alzheimer's disease. Brain Res. 1990;523(2):273–80.
143. McGeer PL, McGeer EG. The amyloid cascade-inflammatory hypothesis of Alzheimer disease: implications for therapy. Acta Neuropathol. 2013;126(4):479–97. https://doi.org/10.1007/s00401-013-1177-7.
144. Petersen RC, Stevens JC, Ganguli M, Tangalos EG, Cummings JL, DeKosky ST. Practice parameter: early detection of dementia: mild cognitive impairment. Neurology. 2001;56(9):1133–42. http://n.neurology.org/content/56/9/1133.abstract.

145. Small GW, Kepe V, Ercoli LM, Siddarth P, Bookheimer SY, Miller KJ, et al. PET of brain amyloid and tau in mild cognitive impairment. N Engl J Med. 2006;355(25):2652–63. https://doi.org/10.1056/NEJMoa054625.
146. Petersen RC. Clinical practice mild cognitive impairment. N Engl J Med. 2011;364:2227–61.
147. Gauthier S, Reisberg B, Zaudig M, Petersen RC, Ritchie K, Broich K, et al. Mild cognitive impairment. Lancet. 2006;367(9518):1262–70. https://www.sciencedirect.com/science/article/pii/S0140673606685425.
148. Reinlieb M, Ercoli LM, Siddarth P, St Cyr N, Lavretsky H. The patterns of cognitive and functional impairment in amnestic and non-amnestic mild cognitive impairment in geriatric depression. Am J Geriatr Psychiatry. 2014;22(12):1487–95. https://www.sciencedirect.com/science/article/pii/S1064748113003989.
149. Yasuno F, Kosaka J, Ota M, Higuchi M, Ito H, Fujimura Y, et al. Increased binding of peripheral benzodiazepine receptor in mild cognitive impairment–dementia converters measured by positron emission tomography with [11C]DAA1106. Psychiatry Res Neuroimaging. 2012;203(1):67–74. http://www.sciencedirect.com/science/article/pii/S0925492711003064.
150. Leng F, Edison P. Neuroinflammation and microglial activation in Alzheimer disease: where do we go from here? Nat Rev Neurol. 2021;17(3):157–72. http://www.nature.com/articles/s41582-020-00435-y.
151. Rogers J, Kirby LC, Hempelman SR, Berry DL, McGeer PL, Kaszniak AW, et al. Clinical trial of indomethacin in Alzheimer's disease. Neurology. 1993;43(8):1609. http://n.neurology.org/content/43/8/1609.abstract.
152. de Jong D, Jansen R, Hoefnagels W, Jellesma-Eggenkamp M, Verbeek M, Borm G, et al. No effect of one-year treatment with indomethacin on Alzheimer's disease progression: a randomized controlled trial. PLoS One. 2008;3(1):e1475. https://pubmed.ncbi.nlm.nih.gov/18213383.
153. Thal LJ, Ferris SH, Kirby L, Block GA, Lines CR, Yuen E, et al. A Randomized, double-blind, study of rofecoxib in patients with mild cognitive impairment. Neuropsychopharmacology. 2005;30(6):1204–15. https://doi.org/10.1038/sj.npp.1300690.
154. Aisen PS, Schafer KA, Grundman M, Pfeiffer E, Sano M, Davis KL, et al. Effects of rofecoxib or naproxen vs placebo on Alzheimer disease progression: a randomized controlled trial. JAMA. 2003;289(21):2819–26. https://doi.org/10.1001/jama.289.21.2819.
155. Scharf S, Mander A, Ugoni A, Vajda F, Christophidis N. A double-blind, placebo-controlled trial of diclofenac/misoprostol in Alzheimer's disease. Neurology. 1999;53(1):197. http://n.neurology.org/content/53/1/197.abstract.
156. Aisen PS, Davis KL, Berg JD, Schafer K, Campbell K, Thomas RG, et al. A randomized controlled trial of prednisone in Alzheimer's disease. Neurology. 2000;54(3):588. http://n.neurology.org/content/54/3/588.abstract.
157. AD2000 Collaborative Group. Aspirin in Alzheimer's disease (AD2000): a randomised open-label trial. Lancet Neurol. 2008;7(1):41–9. http://www.sciencedirect.com/science/article/pii/S1474442207702934.
158. ADAPT Research Group. Alzheimer's disease anti-inflammatory prevention trial: design, methods, and baseline results. Alzheimers Dement. 2009;5(2):93–104. http://www.sciencedirect.com/science/article/pii/S1552526008029841.
159. Breitner JC, Baker LD, Montine TJ, Meinert CL, Lyketsos CG, Ashe KH, et al. Extended results of the Alzheimer's disease anti-inflammatory prevention trial. Alzheimers Dement. 2011;7(4):402–11. https://doi.org/10.1016/j.jalz.2010.12.014.
160. Leoutsakos J-MS, Han D, Mielke MM, Forrester SN, Tschanz JT, Corcoran CD, et al. Effects of general medical health on Alzheimer's progression: the Cache County Dementia Progression Study. Int Psychogeriatr. 2012;24(10):1561–70. https://pubmed.ncbi.nlm.nih.gov/22687143.

161. Hanzel CE, Pichet-Binette A, Pimentel LSB, Iulita MF, Allard S, Ducatenzeiler A, et al. Neuronal driven pre-plaque inflammation in a transgenic rat model of Alzheimer's disease. Neurobiol Aging. 2014;35(10):2249–62.
162. Philippens IH, Ormel PR, Baarends G, Johansson M, Remarque EJ, Doverskog M. Acceleration of amyloidosis by inflammation in the amyloid-beta marmoset monkey model of Alzheimer's disease. J Alzheimers Dis. 2017;55(1):101–13.
163. Fotuhi M, Zandi PP, Hayden KM, Khachaturian AS, Szekely CA, Wengreen H, et al. Better cognitive performance in elderly taking antioxidant vitamins E and C supplements in combination with nonsteroidal anti-inflammatory drugs: the Cache County Study. Alzheimers Dement. 2008;4(3):223–7. https://doi.org/10.1016/j.jalz.2008.01.004.
164. Szekely CA, Breitner JCS, Fitzpatrick AL, Rea TD, Psaty BM, Kuller LH, et al. NSAID use and dementia risk in the Cardiovascular Health Study: role of APOE and NSAID type. Neurology. 2008;70(1):17–24. https://pubmed.ncbi.nlm.nih.gov/18003940.
165. Sweet RA, Seltman H, Emanuel JE, Lopez OL, Becker JT, Bis JC, et al. Effect of Alzheimer's disease risk genes on trajectories of cognitive function in the cardiovascular health study. Am J Psychiatry. 2012;169(9):954–62. https://doi.org/10.1176/appi.ajp.2012.11121815.
166. Pasqualetti P, Bonomini C, Dal Forno G, Paulon L, Sinforiani E, Marra C, et al. A randomized controlled study on effects of ibuprofen on cognitive progression of Alzheimer's disease. Aging Clin Exp Res. 2009;21(2):102–10. https://doi.org/10.1007/BF03325217.
167. Bradburn S, Murgatroyd C, Ray N. Neuroinflammation in mild cognitive impairment and Alzheimer's disease: a meta-analysis. Ageing Res Rev. 2019;50:1–8.
168. Yaqub M, Van Berckel BN, Schuitemaker A, Hinz R, Turkheimer FE, Tomasi G, et al. Optimization of supervised cluster analysis for extracting reference tissue input curves in (R)-[11C] PK11195 brain PET studies. J Cereb Blood Flow Metab. 2012;32(8):1600–8.
169. Spangenberg EE, Green KN. Inflammation in Alzheimer's disease: lessons learned from microglia-depletion models. Brain Behav Immun. 2017;61:1–11.

# A Potential Role for Neuroinflammation in ADHD

15

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**Abstract**

Attention deficit hyperactivity disorder (ADHD) is a neurobehavioural disorder in children and adolescents. Although increases in oxidative stress and disturbances of neurotransmitter system such as the dopaminergic and abnormalities in several brain regions have been demonstrated, the pathophysiology of ADHD is not fully understood. Nevertheless, ADHD involves several factors that have been associated with an increase in neuroinflammation. This chapter presents an overview of factors that may increase neuroinflammation and play a potential role in the development and pathophysiology of ADHD. The altered immune response, polymorphisms in inflammatory-related genes, ADHD comorbidity with autoimmune and inflammatory disorders and prenatal exposure to inflammation are associated with alterations in offspring brain development and are a risk factor; genetic and environmental risk factors that may increase the risk for ADHD and medications can increase neuroinflammation. Evidence of an association between these factors has been an invaluable tool for research on

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inflammation in ADHD. Therefore, evidence studies have made it possible to generate alternative therapeutic interventions using natural products as anti-inflammatories that could have great potential against neuroinflammation in ADHD.

**Keywords**

ADHD · Autoantibodies · Neuroinflammation · Inflammatory disorders · Cytokines · Polymorphisms

## 15.1 Introduction

Attention deficit hyperactivity disorder (ADHD) is a neurobehavioural disorder characterised by a persistent pattern of age-inappropriate inattention and/or hyperactivity-impulsivity, leading to numerous degrees of functional or developmental impairment, resulting in cognitive behavioural, emotional, and social changes that are pervasive in all social settings [1–4].

Because it is a neurobehavioural disorder, it has been difficult to establish a uniform prevalence; however, a prevalence of 5.9% was found in children and adolescents worldwide [4–6]. While a recent meta-analysis published by Song et al., reports that the persistent prevalence in adults (start in childhood) was 2.5%, leading to a prevalence of approximately 50% towards adulthood, data is consistent with several authors [5–8]. Conversely, the prevalence of symptomatic ADHD (regardless of its onset in childhood) is 2.8% worldwide [9]; also, in people aged 50 years or older, a prevalence of 0.02% is estimated worldwide [10]. Several authors have suggested that gender is an important factor in the prevalence and a higher prevalence was repeatedly seen in boys than in girls at a ratio of approximately 2:1; it was also evaluated with predominant criteria for inattention or in hyperactivity/impulsivity, and different proportions related to gender were observed [5, 11]; it is also suggested that the above relationship is difficult to assess, since depending on gender, the response to different situations, such as depression, stress and anxiety can affect the evaluation criteria [2, 12, 13].

## 15.2 Aetiology in ADHD

There is strong evidence for the high heritability of ADHD, estimated to range between 70% and 80% [4, 14, 15]. Genome-wide association study (GWAS) designs allow the analysis of genome-wide DNA variants to obtain data on the association of ADHD with any gene. A recent GWAS meta-analysis identified 12 loci that harbour a DNA variant that increased the risk of suffering ADHD [16], representing approximately 22% of the heritability of the disorder [4]. However, the effect of sizes of individual loci are too small to be clinically relevant, and these findings do not describe which genes are causal [17]. In contrast, molecular studies have suggested

the participation of rare genetic mutations, known as copy number variants (CNV) [17, 18], and it has been discovered that the genomic regions covered by the CNVs associated with ADHD show a significant overlap with CNVs involved in autism and schizophrenia [18]. Moreover, ADHD shares genetic overlap with 43 phenotypes, including insomnia, mortality, educational outcomes, smoking and depressive disorders [16].

Given the aetiological diversity associated with ADHD, numerous risk factors have been described associated with the disorder. They are classified into two main groups: genetic factors, describing a series of candidate genes associated with ADHD and environmental factors, which establish the peri-, pre- and postnatal events that are involved in the development of ADHD. The genetic studies of ADHD have shown that it is highly polygenetic, so its genetic architecture is explained by thousands of common genetic variants, each with a small effect and rare mutations that have a greater effect [16, 18]. Therefore, a single gene is unlikely to be involved in ADHD; rather, it could interact with several different genes. Thus, several studies on ADHD-associated candidate genes mainly involve genes related to the catecholaminergic system, including the dopamine transporter gene (DAT1) [19, 20], the dopamine D4 receptor gene (DRD4) [21, 22], the dopamine D5 receptor gene (DRD5) [21, 23] and lastly, the catechol-*O*-methyltransferase gene [24]. Conversely, genes involved with other neurotransmitter systems have also been described, such as the serotonin receptor 1B (HTR1B) gene [25, 26] and the nicotinic acetylcholine receptor 4 (CHRNA4) gene [27, 28]. In addition, genes related to the glutamatergic system have also been reported, such as the ionotropic glutamate receptor and *N*-methyl-D-aspartate (NMDA) receptor subunit-encoding genes (GRIN2A and GRIN2B) [29, 30]. Similarly, genes involved in the central nervous system (CNS) development have been described; one of the genes widely studied and associated with ADHD is the gene encoding for a regulatory protein of synaptic vesicles called SNAP25, involved in axonal growth and synaptic plasticity and in the coupling and fusion of the synaptic vesicles in the presynaptic neurons, necessary for regulating neurotransmitter release [31, 32]. Moreover, genes involved in immune regulation have also been reported, such as IL-2, IL-6 and tumour necrosis factor-alpha (TNF-α) [33, 34].

Epidemiological studies have described multiple environmental exposures as associated with ADHD, establishing them as putative causal factors for the disorder [4]. It is estimated that between 10% and 40% of the variations associated with ADHD are explained by environmental factors [35]. Among the pre-and perinatal factors, several events have been identified such as maternal stress during pregnancy [35, 36], perinatal vitamin D deficiency [37], maternal exposure to alcohol and tobacco, associating the latter with an increase of approximately two times the risk of suffering ADHD, since it has been established that maternal smoking places the foetus at risk of birth complications; in addition, the nicotinic receptors can modulate the dopaminergic activity, and it has been shown that dopaminergic alteration is involved in the pathophysiology of ADHD [38–40]. Another of the events widely associated with the disorder is the low birth weight (<2500 g) or premature birth (from 33 to <37 weeks of gestation), where children born are small for the

gestational age and present a greater risk of up to three times to be diagnosed for ADHD [35, 40–42]. Besides, complications in pregnancy or childbirth could also influence the risk of getting ADHD by damaging the brain in the early stages of its development [41, 43]. It has also been reported that exposure to environmental toxins such as organophosphates, polychlorinated biphenyls and lead, with medium to low exposures to lead, were associated with the risk to be diagnosed with ADHD [43, 44], and also a risk has been linked to the mother's overweight or obesity before pregnancy [45, 46] and the maternal age, where the children of adolescent mothers (<20 years) are 78% more likely to be diagnosed with ADHD [47].

Regarding the postnatal factors, the main highlights are exposure to artificial food colours and flavours and food or diet supplements [48]. Moreover, social determinants have also been associated, where a set of psychosocial adversity factors could be influencing ADHD development, such as severe marital discord, low social class, large family size, paternal criminality, family dysfunction, child institutional deprivation and the foster home placement [43, 49, 50].

Although genetic and environmental risk factors in ADHD have been described separately, the course of the disorder is likely influenced by how these factors interact and affect the response of an individual to the environment, so to understand the aetiology of ADHD, fully; it is critical to consider how genes and the environment work together to cause the disorder. Therefore, it is suggested that gene–environment interactions (GxE) could be the main mechanism by which environmental factors increase the risk of ADHD since this interaction describes any phenotypic event that is due to interactions between the environment and genes [43, 51]. The presence of GxE interactions has been reported between several genetic variants, mainly between genes associated with the catecholaminergic transmission and environmental factors, such as maternal alcohol and tobacco consumption and the psychosocial adversity, and thus, the results seem to be more consistent for psychosocial factors compared with prenatal factors [51, 52].

## 15.3 Diagnosis of ADHD

There is no gold standard for diagnosing ADHD; it is based on a personalised clinical examination, where the parents or the caregiver and the patient are interviewed to document the criteria of the disorder, questionnaires and standardised tests can be used to capture behavioural and cognition deficiencies and the diagnosis can be very accurate only if it is made by a licensed physician specialising in the subject and using the international standard manual designated by WHO, the *Diagnostic and Statistical Manual of Mental Disorders* (DSM-5) and the latest version of the American Psychiatric Association [4, 5, 53]. Two criteria for the diagnosis are considered: criterion A1 and criterion A2 (Table 15.1).

For children under 17 years old:

- Predominantly inattentive: at least six symptoms of criterion A1 without the presence of A2

**Table 15.1** Diagnostic criteria for ADHD

| At least six symptoms for each criterion <17 years old | |
|---|---|
| At least five symptoms for each criterion ≥17 years old | |
| **A1-inattention symptoms** | **A2-hyperactivity and impulsivity symptoms** |
| Does not pay attention to details and makes mistakes in activities | Is touching with hands or feet or twists in the seat |
| Difficulty maintaining attention on tasks or recreational activities | Gets up in situations where you are expected to remain seated |
| Seems not to listen when spoken to directly | Runs or climbs in situations where it is not appropriate |
| Does not follow instructions and does not finish tasks or activities | Unable to play or quietly engage in recreational activities |
| Difficulty organising tasks and activities | Acts as if it is "driven by an engine" |
| Avoids or dislikes tasks that require sustained mental effort | Speech excessively |
| Loses things needed for tasks or activities | Answer unexpectedly before a question has been concluded |
| Easily is distracted by external stimuli | Difficulty waiting turns |
| Forgetfulness in daily activities | Interrupts or interferes with others |

- Predominantly hyperactivity/impulsivity: at least six symptoms of criterion A2 without the presence of A1
- Combined: at least 12 symptoms of both criteria

For children over 17 years old or more:

- Predominantly inattentive: at least five symptoms of criterion A1 without the presence of A2
- Predominantly hyperactivity/impulsivity: at least five symptoms of criterion A2 without the presence of A1
- Combined: at least ten symptoms of both criteria

They are required to be present in the last 6 months to obtain a diagnosis, and it is required that some of those symptoms are present before the age of 12 years and that have lasted at least the last 6 months, also that the symptoms are present in different environments and affect daily life. Presently, many researchers are trying to develop biological or computerised tests; progress continues in different lines of research such as neuropsychology, neuroimaging, genetics and electroencephalography, among others with which physicians could easily study and evaluate changes, and thus, the diagnosis would be more accurate [3, 5].

## 15.4 Treatment of ADHD

Treatment for ADHD is considered multimodal, since it can consist of pharmacological, behavioural, psychological and psychoeducational therapy, or a combination thereof. In this sense, following the evidence of a neurochemical basis for ADHD, it was found that medications that favour the dopaminergic and/or noradrenergic pathways seem to be necessary for the clinical efficacy of the pharmacological treatments for ADHD [1, 4, 54]. Moreover, choices of treatment approaches are based on the evaluation of the severity of the symptoms, the presence of comorbidities and the preferred periods of the day to alleviate the symptoms. Therefore, medications approved for ADHD treatment include the psychostimulants (considered first-line agents), such as methylphenidate (MPH) and amphetamines, and non-psychostimulants (considered second-line agents), such as atomoxetine (ATX) and α-2 adrenergic receptor agonists (guanfacine and clonidine). Furthermore, several emerging drugs such as glutamatergic agents are increasingly used as treatment, among which are amantadine and memantine, both NMDA glutamate receptor antagonists and modafinil, which, although the mechanism of action is not well established, the data suggest that it exerts effects on the glutamatergic system since it can stimulate glutamate release in the thalamus, the striatum and the hippocampus [1, 3].

The MPH and amphetamine showed similar mechanisms of action since they exert their effects, inhibiting the dopamine and norepinephrine transporters (DAT and NET), thus increasing the extracellular levels of dopamine (DA) and norepinephrine (NE) [55, 56]. It is known that approximately 75–80% of children with ADHD respond efficiently to psychostimulants. The side effects of psychostimulants therapy are headache, anxiety, insomnia, weight loss, agitation, tics and stomach pain, which depend on the dose [57, 58]. The therapeutic use of psychostimulants in ADHD is associated with increased recreational use and increased risk of intentional overdose related to improper use [59, 60]; therefore, psychostimulants use, often for life, has raised several concerns and controversies over the years [61]. Due to lack of response or partial response to psychostimulants, some patients may not take those medications. The clinical advantages of non-psychostimulants over psychostimulant treatments are that there is no potential for abuse, as there are no known effects on drug abuse-related regions of the brain, such as the nucleus accumbens [62, 63]. Although between 10% and 30% of patients do not respond to psychostimulant treatment, non-psychostimulant drugs are generally better tolerated than psychostimulants, and they have lower efficacy and a longer onset of action [64]. ATX acts by selectively inhibiting the NET at the presynaptic level, thus increasing the NE and DA levels in the hippocampus, pre-frontal cortex (PFC) and cerebellum [62, 63, 65, 66]. Common adverse effects include headache, abdominal pain, nausea, diarrhoea, vomiting, decreased appetite, fatigue, dizziness, mood swings and insomnia, where most of the reported effects were mild or moderate and are often seen in the early stages of treatment and tend to decrease significantly over time [3, 63]. Conversely, the α-2 adrenergic receptor agonists, clonidine and guanfacine, are being used mainly as monotherapy or adjuvant therapy in patients

who present a suboptimal response to psychostimulants or ATX. The exact mechanism of action of α-2A agonists in ADHD is unknown; the predominant theory is that these agents directly mimic the effects of NE on α-2A adrenergic receptors in the PFC [67]. Guanfacine is more selective than clonidine at these receptors and may improve treatment efficacy [68]. The pre-synaptic action of clonidine is through the reduction of NE release in the locus coeruleus [69]. Nevertheless, adverse side effects, including dry mouth, nausea, dizziness, constipation, fatigue, variations in blood pressure and cardiovascular actions, have been observed during therapy [70, 71].

## 15.5 Pathophysiology of ADHD

There is strong evidence that structural, functional and neurochemical brain differences are involved in vital cognitive functions, relating to the pathophysiology of ADHD [72–74]. Neuroimaging studies have shown a reduction in global brain volume of 2.5% in patients with ADHD [75]. Moreover, the reductions reported in white matter have been mainly in the splenium of the corpus callosum that extends to the right cingulate and the right sagittal stratum, suggesting problems in the connections between the two hemispheres in regions involved in attention and perception [5, 73]. ADHD has long been listed as a disorder of the PFC. Its connections and the most relevant circuits of the PFC in ADHD are the dorsal frontostriatal (cognitive control), orbitofrontostriatal (reward processing) and fronto-cerebellar (synchronisation), and the dysfunction in these circuits can occur due to deficits in the PFC or problems in the circuits that transmit information to the cortex [73, 76–78]. Another structure widely involved with changes in ADHD is the thalamus. Thus, an altered profile of thalamocortical connectivity has been observed in patients with ADHD, associated with deficits in the processing of information regarding task performance [79]. Furthermore, an anomaly has been seen in the projection fibres that run through the thalamus, ganglia and medulla in ADHD [80]. The cerebellum is another important area related to the cerebral cortex and involved in ADHD. Brain imaging studies have shown the structural abnormalities of the cerebellum in ADHD where a difference in functional connectivity between the cerebellum and the neocortex was demonstrated [81].

Conversely, ADHD has been associated with an imbalance in the dopaminergic and noradrenergic systems, involving them in the pathophysiology of ADHD. Furthermore, the fronto-subcortical circuit associated with ADHD is rich in catecholaminergic signalling [82, 83]. Dopaminergic signalling pathways are crucial for maintaining of physiological processes and play an important role in the neuromodulation of motor control, motivation, reward and cognitive function [84]. ADHD has been associated with dopaminergic dysfunction, particularly with the mesocortical, mesolimbic and nigrostriatal pathways [85, 86], and alterations in those pathways cause deterioration of cognitive abilities. Present-day hypotheses of DA involvement suggest that the core symptoms in ADHD patients stem from DA decrease due to increased DA reuptake [87]. Moreover, it has also been shown that

NE influences the modulation of arousal, state-dependent cognitive processes, motivation, alertness and wakefulness, as well as the neuromodulation of the mechanisms of reward, learning and memory; it also plays a key role in the pathophysiology of ADHD [85]. Studies from patients with ADHD established that they present a deficient transmission of DA and NE that affect the function of the PFC [82]. Additionally, abnormal levels of DAT have been detected in different brain areas of patients with ADHD [2]. As neurotransmitters of the CNS, the catecholamines DA and NE can undergo autoxidation forming reactive oxygen species (ROS) [88, 89]. Therefore, the reaction products formed by the oxidation of catecholamines result in cellular damage, thus contributing to oxidative stress and neuronal death [90, 91].

Recently, there has been an increasing interest in oxidative and nitrosative stress in ADHD and its potential to contribute to the pathophysiology of the disorder. Oxidative stress is defined as the biochemical imbalance caused by the excessive production of ROS and reactive nitrogen species (RSN), which cause oxidative and nitrosative damage to biomolecules and which the antioxidant systems cannot counteract [34, 92–94]. This imbalance can occur because of the malfunctioning of the antioxidant system or as an excessive generation of ROS. It can be caused by several factors such as mitochondrial dysfunction and genetic and environmental factors. Therefore, excessive ROS/RSN levels may damage the integrity of neurons by oxidizing the polyunsaturated fatty acids (PUFAs), producing more ROS that causes oxidative damage of neurons; thus, the neurons that are rich in mitochondria can generate ROS, causing bioenergetic dysregulations, leading to cell death [94, 95]. Oxidative stress could also modify the inflammatory response; therefore, if there is a redox imbalance, the signalling pathways regulating the immune system are changed, producing a dysregulation of the immune response, and on the contrary, if there is a redox balance, the inflammatory response could act as a defence mechanism [96, 97]. In a chronic state of several disorders such as ADHD, oxidative stress can oxidise proteins and lipids and damage the DNA. Thus, oxidative stress in the CNS could also lead to microglia and reactive astrocytes activation and produce chronic neuroinflammation [98]. Therefore, high oxidative stress could activate the secretion of pro-inflammatory chemokines and cytokines and produce a harmful vicious circle [97, 99]. Accordingly, oxidative stress and neuroinflammation are processes that are intricately linked and can coexist. The association of oxidative stress and neuroinflammation as a potential role in the pathophysiology of ADHD could be influenced by the genetic and environmental factors, catecholaminergic dysregulation, an imbalance between oxidants and antioxidant defences, medications used for handling the disorder and as we will see below by multiple immunological factors that could enhance the neuroinflammation and thus increase even more the oxidative stress and inflammation, which could additionally increase or worsen the symptoms of ADHD, resulting in a harmful vicious circle [94].

## 15.6 Neuroinflammation

Inflammation is a physiological process in which the CNS responds to infections, environmental toxins or injuries that affect homeostasis. The inflammatory response in the CNS (brain and spinal cord) is defined as neuroinflammation. Neuroinflammation is at first a protective response, but chronic inflammation is related to the pathogenesis and progression of several psychiatric and mental health disorders [100–102]. The neuroinflammation increases the risk and promotes the progression of neurodegenerative and neurodevelopmental disorders, including ADHD, through different mechanisms such as glial cell activation, increased oxidative stress, loss of neuronal function and neurodevelopment changes [34, 94].

Glial cells can produce different inflammatory mediators (cytokines, chemokines, ROS, RNS, prostaglandins, leukotrienes and growth factors) in response and depending on the degree of CNS injury [103–105]. Pro-inflammatory cytokines and chemokines trigger activation of surrounding stromal cells, induce glutamate release (excitotoxicity) and increase the permeability of the blood-brain barrier (BBB), allowing more immune cell infiltration in the brain parenchyma, enhancing the inflammatory response [106–109]. Inflammation resolution is performed when the tissue has been repaired and homeostasis is restored and mediated by the release of anti-inflammatory cytokines (IL-10, IL-4, TGF-β and IL-37), lipoxins, resolvins and neuroprotectins [110–112].

Microglia are the resident macrophages of the CNS, originating from myeloid precursor cells in the yolk sac during embryonic development and represent 10% of the CNS cell population [113, 114]. Microglia has pleiotropic functions during CNS development, such as axon guidance, neurite growth, synapse function and plasticity [115]. Microglia activation and accumulation (known as microgliosis) in response to various external and internal stimuli, induce morphological (amoeboid shape) and functional changes (inflammatory mediators production, tissue repair and phagocytosis) [116]. Depending on the detected insult and the microenvironment, microglia can acquire a pro-inflammatory phenotype, called M1. The production of cytokines characterises the M1-phenotype as IL-1β and TNF-α, chemokines, ROS, nitric oxide and prostaglandins. The anti-inflammatory M2-phenotype is characterised by the expression of IL-10, IL-4, TGF-β (transforming growth factor-β), IGF-1 (insulin-like growth factor 1), arginase and other factors [116, 117].

Astrocytes represent about 40% of all brain cells and play a critical role in providing nutrients to neurons, synapse formation and synaptic transmission [118]. Similar to microglia, astrocytes have pro-inflammatory and anti-inflammatory functions depending on the damage. The active state of astrocytes (reactive astrocytes) and their accumulation (astrocytosis or astrogliosis) are a hallmark of neurodegeneration and neuroinflammation. Reactive astrocytes show morphological changes and altered expression proteins, such as glial fibrillary acidic protein, vimentin and glutamine synthetase [118]. Activated microglia and astrocytes are primarily responsible for the productions of ROS and lead to oxidative stress. As indicated above, oxidative stress can induce chronic neuroinflammation and contribute to neurodegeneration [98]. In pathological states, astrocyte activation leads to

astrocyte hypertrophy, proliferation, production of inflammatory mediators and an altered communication between astrocytes and neurons contributing to neuronal damage [118, 119].

Oligodendrocytes are glial cells in the CNS that produce myelin structure that wraps around axons, allows proper conduction of action potentials and provides metabolic support to neurons [120]. Oligodendrocytes express various immunomodulatory molecules such as IL-1β, IL-6, IL-17A, chemokines, tetraspanins, major histocompatibility complex (MHC) proteins, co-stimulatory molecules and proteins of complement. Thus, immunologically active oligodendrocytes can be an important factor in the initiating inflammation or its resolution, especially in demyelinating diseases [121].

The T and B cells are constituents of the adaptive immune system; their activation is antigen-specific. Brain parenchyma under physiological conditions does not contain lymphocytes, but B and T cells reside in the meninges and choroid plexus and can influence brain development and function [122]. B cells have different effector functions depending on the signal receiving and intensity. B cells primary function is to produce antibodies; also, they are antigen-presenting cells (activation T cells), activate inflammatory macrophages and inhibit regulatory immune cells [122, 123]. Through their T cell receptor (TCR), T cells detect antigens that have been presented on MHC molecules by other cells. CD8+ T cells subsets detect antigens presented on MHC class I molecules, and CD4+ T cells detect antigens presented on MHC II molecules. After activation, CD4+ T cells proliferate and differentiate into numerous subsets, including type 1 T helper (Th1) cells, Th2, Th17 and regulatory T cells (Treg) [124]. Meningeal T cells can produce cytokines, neurotransmitters such as γ-aminobutyric acid (GABA), neuromodulators such as serotonin and growth factors (BDNF), which influences neuronal function in physiological and pathological conditions [122, 125]. For example, it has been shown that T cells are responsible for social and cognitive behaviours in mice [126, 127].

Mast cells are effector immune cells resident in several brain areas and are in the meninges, parenchyma of the thalamic hypothalamic region and in the abluminal side of the blood vessels, where they modulate the interaction between meninges and the immune system. Activated mast cells produce IL-6, TNF-α, tryptase, histamine, chymase, corticotrophin-releasing hormone (CRH) and neurotransmitters. They also produce chemokines that can recruit other immune peripheral cells in the brain tissue. Thus, mast cells can exacerbate the development of pathologies affecting the CNS by producing inflammatory cytokines, cytotoxicity and neuronal and glial cell death [128, 129].

### 15.6.1 Key Inflammatory Cytokines

Interleukin-1 (IL-1) are a family of pro-inflammatory cytokines, consisting of 11 members, including IL-1α, IL-1β, IL-1 receptor antagonist (IL-1Ra) and IL-1 receptor accessory proteins (IL-1RAcP), IL-33 and IL-37, and the only anti-inflammatory cytokine of family. IL-1β can trigger inflammatory mediators like

other cytokines and chemokines (as IL-6, TNF-α and IL-8) and can stimulate macrophages, neutrophils, lymphocytes and perivascular endothelial cells, among other cells. IL-1Ra is a regulatory molecule that competes for receptor binding with IL-1a and IL-1β, blocking their role in immune activation. In different brain regions, various components from the IL-1 family are constitutively expressed in healthy individuals. IL-1β is a pivotal mediator and has been shown to influence dopaminergic and noradrenergic function, feeding, fever, sickness behaviour and sleep [128, 130–132]. During an event of injury or infection, microglia is the primary source of IL-1β. Microglia, through the inflammasome, generates active caspase-1 that cleaves pro-IL-1β to convert it to active IL-1β. IL-1β activation induces a cascade of events, leading to activation of nuclear transcription factor-κB (NF-κB) and transcription of genes involved in the immune response. In addition, IL-1-β-activated microglia produces proteases, ROS, RNS, prostaglandins, cytokines and chemokines. All these mediators are central to the development of neuropsychological, neurodegenerative and demyelinating diseases [110, 128, 133, 134].

TNF-α is a pro-inflammatory cytokine, expressed under physiologic conditions by microglia and neurons, but its expression increases in activated microglia, neurons, oligodendrocytes, reactive astrocytes, endothelial cells and ependymal cells in brain injuries and chronic diseases. TNF-α controls numerous physiological processes in the CNS, at low concentrations, promoting neurogenesis, axonogenesis and synaptic plasticity [130, 135]. TNF-α mediates different physiological and pathological functions, by activating types 1 and 2 TNF receptors (TNFR1 and TNFR2). The binding of TNF-α with TNFR1 leads to the recruitment of complex 1, which allows nuclear translocation of transcription factors, as NF-κB and AP1 and transcription of pro-inflammatory mediators and anti-apoptotic proteins. However, an alternative pathway of TNRF1 is mediated by complex II recruitment that can turn on apoptosis. Conversely, TNFR2, similar to TNFR1, leads to NF-κB activation and promotes cell survival, resolution of inflammation and tissue repair [136–139].

The interleukin-6 (IL-6) is a crucial mediator in regulating the inflammatory response and can have both pro- and anti-inflammatory activity. The anti-inflammatory effects of IL-6 are through the activation of IL-1ra and IL-10 and the decrease of TNF-α and IL-1. IL-6 is widely produced by many CNS cells, but IL-6 receptor-α (IL-6R), essential for the cellular response, is differentially expressed in cells [140]. IL-6 binds to membranal IL-6R, which triggers oligomerisation with gp130, and downstream signalling culminates in signal transducers and activators of transcription 3 (STAT3) activation, which in turn mediated expression of IL-6-regulated genes. But cells that lack the IL-6R in the CNS can respond to IL-6 using an alternate mode of signalling (called trans-signalling) through soluble IL-6R (sIL-6R), which retains its biological activity and can activate signalling through gp130 expressed in cells lacking IL-6R [141]. IL-6 is essential in the CNS development and the appropriate functioning of neurons and glial cells. Neurons, microglia and endothelial cells produce IL-6, but astrocytes are the main source. IL-6 dysregulation can lead to inflammatory, autoimmune and psychiatric disorders [142].

Interleukin-10 (IL-10) is an anti-inflammatory cytokine that negatively regulates inflammation. IL-10 is expressed in the brain; specifically, it is produced by microglia, astrocytes and neurons. IL-10 binds to the cell surface receptor, a heterotetrameric complex IL-10R1/IL-10R2; this interaction leads to the activation of downstream signalling cascades including the STAT3 and AKT pathways. IL-10 plays an important role in inhibiting the production of pro-inflammatory mediators, decreasing cytokine receptor and MHC class II expression, neuroprotection and modulation of synaptic activity. Moreover, IL-10 regulates GABAergic transmission in the hippocampal neurons via pre-and post-synaptic mechanisms [142, 143].

## 15.7 Neuroinflammation in ADHD

Pro-inflammatory cytokines are closely involved in the onset, development and symptoms of ADHD (Table 15.2). Patients with ADHD have elevated concentrations of pro-inflammatory cytokines (as IL-6 and TNF-α) and reduced levels of anti-inflammatory cytokines (IL-4, IL-2 and IFN-γ) and brain-derived neurotrophic factor (BDNF) [144, 147–149]. Thus, high concentrations of cytokines and chemokines and oxidative stress markers were found in the serum of juvenile spontaneously hypertensive rats (SHRs, used as a rodent model for ADHD) compared with the age-matched controls [150]. Interestingly, there was an association between high pro-inflammatory cytokines levels and severity of symptoms in children with ADHD [151–153]. However, this correlation was lost in the adult patients with ADHD [154, 158]. Several studies have shown that IL-6 may be a hallmark of ADHD pathogenesis since increased IL-6 alters attention and memory through its effects on synaptic plasticity in the hippocampus and PFC [144]. Moreover, it was demonstrated that IL-6 inhibited the neurogenesis in the hippocampus through the blockade of the differentiation of neural progenitor cells, which compromised brain development and increased the risk of ADHD [145, 159]. However, there are also contrasting results because it was found that the IL-6 levels did not correlate with ADHD symptoms severity. Nevertheless, findings from numerous studies suggested that the increased IL-6 levels can be directly related to the ADHD aetiology [148]. Metalloproteinases (MMPs) are proteolytic enzymes involved in the cleave of components in the extracellular matrix, adhesion molecules and cytokines and growth factors [160]. Particularly, the MMPs are involved in the memory and the learning process in the brain. Therefore, in ADHD patients, it was found that there was a correlation between MMP-2 and MMP-9 levels with cognitive problems, as well as a negative correlation between MMP-2, MMP-9 and TNF-α levels with the intelligence quotient (IQ) [146]. An overview of the factors involved in neuroinflammation is shown in Fig. 15.1.

**Table 15.2** Principal neuroinflammatory mediators associated with ADHD

| Inflammatory factors | Main findings | References |
|---|---|---|
| IL-6 | High levels in children with ADHD compared with control and affects attention and memory by its effect on neurogenesis and synaptic plasticity in the hippocampus and PFC | [144, 145] |
| TNF-α, MMP-2, MMP-9 | Higher concentrations in patients with ADHD and positive correlation with age and a negative correlation with IQ and also, correlation with cognitive and inattention problems | [146] |
| IL-6, TNF-α | In patients with ADHD and obesity, it was found a correlation between the severity of hyperactivity/impulsivity | [147] |
| IL-6, TNF-α | Higher serum levels in children with ADHD, did not correlate with IQ or ADHD symptoms severity | [148] |
| CRP, IL-6, TNF-α, BDNF | Higher plasma levels (CRP and IL-6) | [149] |
| | Lower levels of TNF-α and BDNF in ADHD young | |
| IL-1β, IL-6, TNF-α, TGF-β, MCP-1, RANTES, IP-10 | There were significant increases of serum and/or tissue in the ADHD animal model when compared with controls | [150] |
| IL-8, TNF-α, VEGF, MMP-9 | Children born preterm express high concentrations Correlation with the severity of ADHD symptoms | [151] |
| IL-6, IL-8, ICAM-3, VEGF-R1, VEGF-R2, TNF-RI | Children born prematurely with increases in blood during the first postnatal weeks are associated with an attention problem | [152] |
| IL-13, IL-16, IL-2 | High concentrations in serum were associated with hyperactivity, inattention, and opposition | [153] |
| IL-6, IL-10, anti-Yo antibodies | High cytokine and antibodies levels in children with ADHD compared with controls | [154] |
| IL-2, IL-6, TNF-α, IL-16, S100B | Gene polymorphisms are highly associated with ADHD | [155, 156] |
| Anti-DAT antibodies | These antibodies are present in patients with ADHD and are noteworthy as a sign of inflammation | [157] |

### 15.7.1 Genetic Variants in ADHD

ADHD is a highly inherited disorder, and there is a lot interest in identifying the genetic factors involved. Some genetic risk factors for inattention and hyperactivity/impulsivity are shared with other disorders, and others are unique to ADHD [161]. Different GWAS studies have identified genetic variants associated with an increased risk for ADHD [162, 163]. There are polymorphisms in genes related to inflammatory pathways, such as angiogenic (NRP1 and NRP2), neurotrophic (NTRK1 and NTRK3), cytokine (IL-16 and S100B) and kynurenine (CCBL1 and CCBL2) genes, associated with ADHD [155]. IL-1RA is a molecule that competes for receptor binding with IL-1a and IL-1b, blocking their role in immune activation.

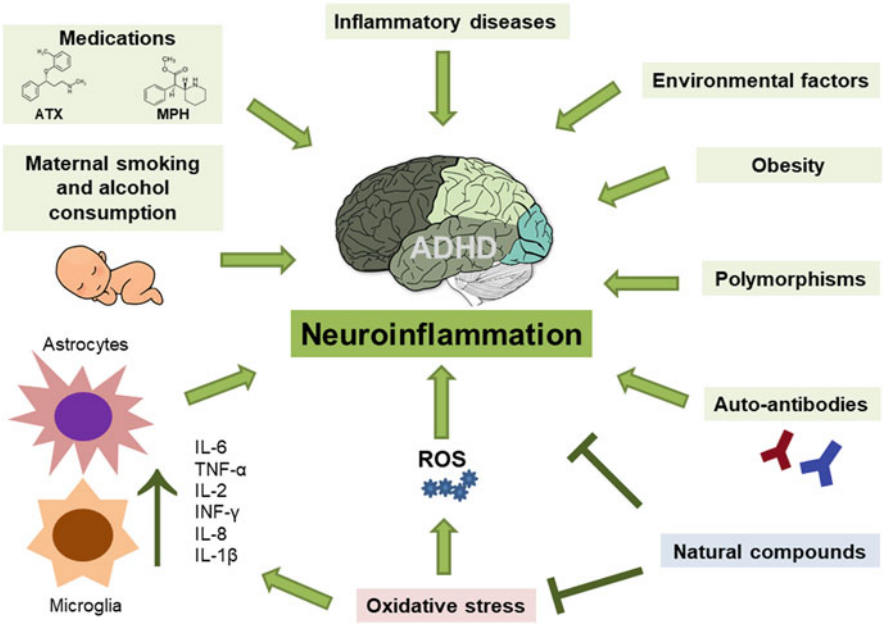


**Fig. 15.1** Potential role of neuroinflammation in the pathophysiology of ADHD. Comorbidity between ADHD and several autoimmune and inflammatory disorders, environmental and genetic factors, medications used for the treatment, maternal alcohol consumption and smoking, obesity, some polymorphisms, and also oxidative stress may increase the risk of neuroinflammation in ADHD and natural compounds may have anti-inflammatory and antioxidant effects against neuroinflammation in ADHD

Polymorphisms in the IL-1Ra gene, IL1-RN, also have been directly associated with ADHD [131]. Other studies involve neurotrophic factors (NTFs), molecules that control survival, differentiation and neural functions. Polymorphisms in NTFs and their receptors might be involved in the genetic predisposition to ADHD. Thus, it has been shown the contribution of the ciliary neurotrophic factor receptor (CNTFR) locus as a predisposition factor to childhood and adulthood in ADHD. At the same time, variants of NTF3 and NTRK2 are childhood-specific [156].

### 15.7.2 Autoantibodies in ADHD

A disruption of BBB integrity due to an inflammatory process in the brain leads to increased flux of CNS antigens to peripheral lymphoid organs, with a subsequent autoimmune response (autoantibodies production). Therefore, elevated levels of anti-basal ganglia antibodies and antibodies against the DAT in ADHD with notable signs of inflammation have been found [157, 164]. In addition, it was demonstrated that anti-Yo antibodies (Purkinje cell antibodies) were associated with high cytokine serum levels in children with ADHD compared with controls [154, 165]. However,

more studies are needed to establish a possible role of autoantibodies in ADHD pathogenesis.

### 15.7.3 Comorbidity of ADHD with Other Diseases and Factors

Numerous studies have shown a relationship between ADHD, inflammatory and autoimmune diseases. The systemic increase in pro-inflammatory cytokines levels due to common mechanisms (genetic and/or environmental) could cause neuroinflammation and impaired cognitive functions, resulting in the onset and development of neurodevelopmental disorders [166]. Furthermore, inflammatory cytokines can disrupt the maturation of PFC regions and the dopaminergic systems involved in ADHD pathology [167].

Atopic eczema, rhinitis and allergic asthma are among the most common chronic diseases worldwide and are grouped as atopic diseases. Multiple studies suggest that in children with ADHD are more likely to develop [168–170]. Moreover, children with atopic disease have increased levels of pro-inflammatory cytokines that pass through the BBB and affect mechanisms involved in the behaviour and emotion [171, 172]. Children (age 8–9) with asthma had a twofold greater risk of having one or more symptoms of hyperactivity/impulsivity and a more than twofold risk of having three or more symptoms among the older age (13–14 years), and these results were independent of asthma medications [169].

Obesity is also a risk factor for the onset of neuroinflammation, and a chronic and low-grade inflammatory process is present in adipose tissue in obese individuals. Therefore, adipocytes can also release many pro-inflammatory mediators that increase the concentration of peripheral cytokines and predispose them to neuroinflammation [173]. Thus, multiple studies suggest that obesity may increase inflammation and contribute to ADHD symptoms [147, 174]. For example, in patients with obesity and ADHD, a correlation was found between the hyperactivity/impulsivity scores and cytokines IL-6 and TNF-α, which held after controlling for body mass index and oppositional symptoms [147]. Interestingly, obesity risk allele polymorphisms are associated with ADHD and related to some symptoms such as inattention and hyperactivity/impulsivity. For example, one of the genes was the cell adhesion molecule 2, which was associated with hyperactivity [175]. Furthermore, maternal obesity has also been associated with inflammation and behavioural and cognitive alterations in offspring linked to neurodevelopmental disorders such as ADHD and autism [176].

Genetic, epigenetic and environmental interactions have a principal role in brain formation and function. Thus, pre-clinical studies show that environmental disruptions to the developing CNS in early life can result in alterations in the neurobehavioural, cognitive and mental health of individuals [100, 177]. In this manner, several findings show exposure to prenatal inflammation associated with behavioural symptoms related to ADHD such as inattention, hyperactivity, impulsivity and impaired learning and memory [100]. Moreover, the effect of maternal inflammation on the neurodevelopment of the offspring is multifactorial and is

generally related to other perinatal complications such as preterm delivery, low birth weight and placental ischaemia [178, 179]. Children with ADHD whose mothers were exposed to moderate and severe stress during pregnancy develop more severe symptoms than offspring with ADHD whose mothers were not exposed to prenatal stress [180]. Additionally, children born premature and whose mothers suffered from stress during pregnancy had a significantly increased risk of developing asthma [181].

Alcohol exposure is related to neurodegeneration and cognitive dysfunction resulting from microglial activation and inflammatory response [182]. Any amount of alcohol exposure during prenatal development can increase the risk of developing cognitive or psychiatric disorders; even low levels of foetal alcohol exposure can negatively affect cognitive function in the offspring [183]. Thus, alcohol-induced inflammation could contribute to the development of ADHD through several mechanisms such as inhibition of neurogenesis, T-cell infiltration through the BBB and the pro-neuronal survival transcription factor CREB [145]. Conversely, it has also been shown that smoking during pregnancy is associated with ADHD. A meta-analysis study showed a dose-response relationship between smoking during pregnancy and the risk of ADHD in offspring and specifically with a more severe presentation of ADHD symptoms, including comorbidity with behaviour disorders [184, 185]. Besides, prenatal nicotine exposure in mice produced hyperactivity similar to the human ADHD phenotype, decreased DA turnover in the frontal cortex, decreased cortical volume and radial thickness. However, MPH administration decreased the hyperactivity and increased the DA turnover in the frontal cortex [186]. Moreover, it was found by quantifying the concentration of cotinine levels, used as a biomarker indicating nicotine exposure and revealed an association with a dose-response and nicotine exposure during pregnancy and offspring with ADHD [39]. Furthermore, smoking during pregnancy could have consequences such as preterm delivery, low birth weight, placenta abruption, development of obesity or overweight, reduced foetal lung development and increased infection [187, 188]. Several studies have also shown a connection between prenatal exposure to different infectious agents (bacteria and virus) and the risk of ADHD in the offspring [189, 190]. Also, neonatal infection associated with systemic inflammatory responses during the postnatal stage was associated with the risk of ADHD [151]. In this context, a recent study showed that preterm infants who had neonatal infection had an increased risk of severe motor impairment in ADHD and IQ delay than those without infection and who had preterm births [191]. Conversely, long-term maternal use of acetaminophen during pregnancy was associated with ADHD in offspring [192]. Acetaminophen is the most commonly used medication for analgesic and antipyretic purposes among mothers during pregnancy and infants in early life. Moreover, it was shown that prenatal exposure to acetaminophen was associated with an increased ADHD risk in offspring, regardless of gestational infections or maternal mental health diseases [193]. But contradictory results have been reported, where it was found that maternal fever in the first trimester can be a risk factor for ADHD (mainly inattention). However, this risk factor was independent of the use of acetaminophen [194].

There is now evidence that exposure to pollutants (including industrial chemicals, pesticides, heavy metals and phytoestrogens) during early gestational stages can increase the risk of ADHD through inflammatory mechanisms [195]. Through maternal immune activation, studies in animal models have generated information on neurodevelopmental disorders compatible with ADHD [196]. Thus, prenatal exposure to an inflammatory environment can be associated with changes in the development of the brain in the foetus, including anatomical changes, such as reduction in the volume of the areas in the cortical zone, observed in patients with ADHD [100]. Nowadays, there is a correlation between poor mental development and the presence of pesticides during the human gestational stage. Nevertheless, there are few association studies between pesticides and ADHD [197]. Studies in animal models, have established certain biological correlations on pesticide exposure with neurodevelopmental and behaviour alterations, but are not conclusive [198]. Bisphenol A is a ubiquitous endocrine-disrupting chemical associated with disturbances in neurobehavioural development [199]. Although, overall prenatal exposure to bisphenol A can contribute to neurobehavioural outcomes in children, the evidence is still limited; however, ADHD symptoms constantly suggested association with both prenatal and concurrent exposure to bisphenol A [200, 201].

Epidemiologic studies have displayed a close correlation between exposure to phthalates and a disturbance during neurodevelopment with ADHD [202]. However, there are inconsistencies between the presence of metabolites and the neurobehavioural assessment criteria, possibly due to heterogeneity in neurodevelopmental tests [203]. In addition, perfluoroalkyl and polyfluoroalkyl substances (PFASs) are persistent pollutants that can have neurotoxic effects because they can cross the placental barrier. However, current evidence is insufficient to establish the association between prenatal exposure to PFASs and ADHD symptoms or cognitive dysfunctions in preschool children [204, 205]. Finally, both human and animal studies showed a critical role for the placenta in mediating the impact of chronic inflammatory state, foetal immune activation and increased oxidative stress on foetal brain development [206]. Therefore, future research should focus on further elucidating the mechanism of toxicity of pollutants like pesticides on the axis foetal brain/placental and the possible consequences on foetal brain development in ADHD.

## 15.8 ADHD Medications and Neuroinflammation

Several studies have shown that treatment with MPH can increase neuroinflammation; thus, the highest concentration and chronic administration of MPH resulted in microglial activation in multiple brain regions of rats [207]. Also, it was demonstrated that MPH induced DA neuron loss and inflammation through the increase in mRNA levels of pro-inflammatory cytokines (IL-6 and TNF-α) in the mice striatum [208]. Moreover, MPH treatment improved cell survival at low concentrations in monocytic immune cell lines, and conversely, highest concentrations caused a significant reduction in monocytic cell survival

[209]. Recently, the dose-dependent administration of high doses of MPH altered motor activity, anxiety and increased TNF-α and IL-1β levels, lipid peroxidation and glutathione oxidised levels in the hippocampus and cerebral cortex of adult rats [210]. In contrast, acute treatment with ATX decreased the expression of microglial activation markers (CD40 and CD11b) and IL-1β, TNF-α and iNOS in the cerebral cortex of rats after systemic treatment with lipopolysaccharides [211]. ATX also protected against ischaemic damage, attenuating the activation of astrocytes and microglia in the ischaemic hippocampal region in the gerbil [212]. Further studies are necessary to investigate the side effects caused by medications used for ADHD treatment (mainly on the inflammatory process) during critical periods of neurodevelopment.

## 15.9 Use of Dietary and Natural Compounds Against Neuroinflammation in ADHD

Every day, more studies focus on the neuroprotective effects of dietary or natural products against oxidative stress and/or inflammation because they may be alternative therapeutic interventions with fewer side effects and better tolerated to improve symptoms of ADHD. Several dietary or natural components have been studied in patients due to the therapeutic benefits in ADHD, focused on its anti-inflammatory and/or antioxidant activities such as diverse flavonoids that have anti-inflammatory activities and include several natural polyphenols that are found in a great quantity in vegetables, fruits, red wine and green tea [58, 213, 214]. Quercetin, a flavonoid compound present in cilantro, onion, asparagus, capers, red leaf lettuce, lovage, dill, berries and apples may exert anti-inflammatory properties [215]. *N*-Acetylcysteine, a precursor of the antioxidant glutathione, may also exert anti-inflammatory activities and is found in the onion [34, 214]. Omega-3 fatty acids have anti-inflammatory activities and the two principals are eicosapentaenoic acid and docosahexaenoic acid and are found mainly in oily fish [34, 213, 214, 216]. Sulforaphane may exert anti-inflammatory activities and is found in the highest concentrations in cauliflower and broccoli [34]. There are other compounds such as ginseng, St. John's wort, *Ginkgo biloba* and passion flower [58] used with ADHD patients against oxidative stress; however, it is not ruled out that they may also have anti-inflammatory effects; nevertheless, further studies are necessary to confirm such effect. Accordingly, these natural compounds could improve ADHD progression due to their anti-inflammatory and/or antioxidant properties.

## 15.10 Conclusion

The pathophysiology of ADHD has been associated with an increase in neuroinflammation. Therefore, several of the immune factors discussed in this chapter appear to play a potential role in the pathological process of ADHD. Factors such as an altered immune response, genetic associations and also exposure to

environmental factors, such as pollutant exposure, maternal smoking and alcohol consumption, could trigger inflammation during the early postnatal period and childhood and, thus, influence risk for ADHD; comorbidity between ADHD and several autoimmune and inflammatory disorders, some medications used for the treatment and also some polymorphisms in genes can cause alterations of the inflammatory response in ADHD. Conversely, dietary and natural compounds with potent anti-inflammatory and antioxidant properties could improve the inflammation and reduce oxidative stress and be used as alternative therapeutic interventions for ADHD. In summary, the evidence indicates a potential role of neuroinflammation in the pathophysiology of ADHD. However, the new tools and technologies will enable future research to evaluate inflammation in patients with ADHD in a non-invasive aspect and, therefore, have more reliable results. Further clinical trials and future, well-designed studies are still needed to confirm the potential role of neuroinflammation in ADHD.

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## References

1. Cortese S. Pharmacologic treatment of attention deficit-hyperactivity disorder. N Engl J Med. 2020;383(11):1050–6.
2. Faraone SV, Asherson P, Banaschewski T, Biederman J, Buitelaar JK, Ramos-Quiroga JA, et al. Attention-deficit/hyperactivity disorder. Nat Rev Dis Prim. 2015;1:15020.
3. Corona JC. Pharmacological approaches for the treatment of attention-deficit/hyperactivity disorder. In: Kyser BM, editor. Attention-deficit hyperactivity disorder: diagnosis, prevalence and treatment. New York, NY: Nova Science Publishers, Inc.; 2021. p. 1–39.
4. Posner J, Polanczyk GV, Sonuga-Barke E. Attention-deficit hyperactivity disorder. Lancet. 2020;395(10222):450–62.
5. Faraone SV, Banaschewski T, Coghill D, Zheng Y, Biederman J, Bellgrove MA, et al. The World Federation of ADHD International Consensus Statement: 208 Evidence-based conclusions about the disorder. Neurosci Biobehav Rev. 2021;128:789–818.
6. Polanczyk GV, Willcutt EG, Salum GA, Kieling C, Rohde LA. ADHD prevalence estimates across three decades: an updated systematic review and meta-regression analysis. Int J Epidemiol. 2014;43(2):434–42.
7. Simon V, Czobor P, Balint S, Meszaros A, Bitter I. Prevalence and correlates of adult attention-deficit hyperactivity disorder: meta-analysis. Br J Psychiatry. 2009;194(3):204–11.
8. Song P, Zha M, Yang Q, Zhang Y, Li X, Rudan I. The prevalence of adult attention-deficit hyperactivity disorder: a global systematic review and meta-analysis. J Glob Health. 2021;11:04009.
9. Fayyad J, Sampson NA, Hwang I, Adamowski T, Aguilar-Gaxiola S, Al-Hamzawi A, et al. The descriptive epidemiology of DSM-IV Adult ADHD in the World Health Organization World Mental Health Surveys. Attent Deficit Hyperact Disord. 2017;9(1):47–65.
10. Dobrosavljevic M, Solares C, Cortese S, Andershed H, Larsson H. Prevalence of attention-deficit/hyperactivity disorder in older adults: a systematic review and meta-analysis. Neurosci Biobehav Rev. 2020;118:282–9.
11. Sayal K, Prasad V, Daley D, Ford T, Coghill D. ADHD in children and young people: prevalence, care pathways, and service provision. Lancet Psychiatry. 2018;5(2):175–86.

12. Willcutt EG, Nigg JT, Pennington BF, Solanto MV, Rohde LA, Tannock R, et al. Validity of DSM-IV attention deficit/hyperactivity disorder symptom dimensions and subtypes. J Abnorm Psychol. 2012;121(4):991–1010.
13. Polanczyk G, de Lima MS, Horta BL, Biederman J, Rohde LA. The worldwide prevalence of ADHD: a systematic review and metaregression analysis. Am J Psychiatry. 2007;164(6): 942–8.
14. Faraone SV, Perlis RH, Doyle AE, Smoller JW, Goralnick JJ, Holmgren MA, et al. Molecular genetics of attention-deficit/hyperactivity disorder. Biol Psychiatry. 2005;57(11):1313–23.
15. Purper-Ouakil D, Ramoz N, Lepagnol-Bestel AM, Gorwood P, Simonneau M. Neurobiology of attention deficit/hyperactivity disorder. Pediatr Res. 2011;69(5 Pt 2):69R–76R.
16. Demontis D, Walters RK, Martin J, Mattheisen M, Als TD, Agerbo E, et al. Discovery of the first genome-wide significant risk loci for attention deficit/hyperactivity disorder. Nat Genet. 2019;51(1):63–75.
17. Nigg JT, Sibley MH, Thapar A, Karalunas SL. Development of ADHD: etiology, heterogeneity, and early life course. Annu Rev Dev Psychol. 2020;2(1):559–83.
18. Thapar A. Discoveries on the genetics of ADHD in the 21st century: new findings and their implications. Am J Psychiatry. 2018;175(10):943–50.
19. Grunblatt E, Werling AM, Roth A, Romanos M, Walitza S. Association study and a systematic meta-analysis of the VNTR polymorphism in the 3′-UTR of dopamine transporter gene and attention-deficit hyperactivity disorder. J Neural Transm (Vienna). 2019;126(4):517–29.
20. Kopeckova M, Paclt I, Petrasek J, Pacltova D, Malikova M, Zagatova V. Some ADHD polymorphisms (in genes DAT1, DRD2, DRD3, DBH, 5-HTT) in case-control study of 100 subjects 6-10 age. Neuro Endocrinol Lett. 2008;29(2):246–51.
21. Gizer IR, Ficks C, Waldman ID. Candidate gene studies of ADHD: a meta-analytic review. Hum Genet. 2009;126(1):51–90.
22. Sunohara GA, Roberts W, Malone M, Schachar RJ, Tannock R, Basile VS, et al. Linkage of the dopamine D4 receptor gene and attention-deficit/hyperactivity disorder. J Am Acad Child Adolesc Psychiatry. 2000;39(12):1537–42.
23. Lowe N, Kirley A, Hawi Z, Sham P, Wickham H, Kratochvil CJ, et al. Joint analysis of the DRD5 marker concludes association with attention-deficit/hyperactivity disorder confined to the predominantly inattentive and combined subtypes. Am J Hum Genet. 2004;74(2):348–56.
24. Biederman J, Kim JW, Doyle AE, Mick E, Fagerness J, Smoller JW, et al. Sexually dimorphic effects of four genes (COMT, SLC6A2, MAOA, SLC6A4) in genetic associations of ADHD: a preliminary study. Am J Med Genet B Neuropsychiatr Genet. 2008;147B(8):1511–8.
25. Ickowicz A, Feng Y, Wigg K, Quist J, Pathare T, Roberts W, et al. The serotonin receptor HTR1B: gene polymorphisms in attention deficit hyperactivity disorder. Am J Med Genet B Neuropsychiatr Genet. 2007;144B(1):121–5.
26. Ribases M, Ramos-Quiroga JA, Hervas A, Bosch R, Bielsa A, Gastaminza X, et al. Exploration of 19 serotoninergic candidate genes in adults and children with attention-deficit/hyperactivity disorder identifies association for 5HT2A, DDC and MAOB. Mol Psychiatry. 2009;14(1):71–85.
27. Lee J, Laurin N, Crosbie J, Ickowicz A, Pathare T, Malone M, et al. Association study of the nicotinic acetylcholine receptor alpha4 subunit gene, CHRNA4, in attention-deficit hyperactivity disorder. Genes Brain Behav. 2008;7(1):53–60.
28. Wallis D, Arcos-Burgos M, Jain M, Castellanos FX, Palacio JD, Pineda D, et al. Polymorphisms in the neural nicotinic acetylcholine receptor alpha4 subunit (CHRNA4) are associated with ADHD in a genetic isolate. Attent Deficit Hyperact Disord. 2009;1(1):19–24.
29. Huang X, Wang M, Zhang Q, Chen X, Wu J. The role of glutamate receptors in attention-deficit/hyperactivity disorder: from physiology to disease. Am J Med Genet B Neuropsychiatr Genet. 2019;180(4):272–86.
30. Kim JI, Kim JW, Park S, Hong SB, Lee DS, Paek SH, et al. The GRIN2B and GRIN2A gene variants are associated with continuous performance test variables in ADHD. J Atten Disord. 2020;24(11):1538–46.

31. Feng Y, Crosbie J, Wigg K, Pathare T, Ickowicz A, Schachar R, et al. The SNAP25 gene as a susceptibility gene contributing to attention-deficit hyperactivity disorder. Mol Psychiatry. 2005;10(11):998–1005, 973.
32. Guan L, Wang B, Chen Y, Yang L, Li J, Qian Q, et al. A high-density single-nucleotide polymorphism screen of 23 candidate genes in attention deficit hyperactivity disorder: suggesting multiple susceptibility genes among Chinese Han population. Mol Psychiatry. 2009;14(5):546–54.
33. Drtilkova I, Sery O, Theiner P, Uhrova A, Zackova M, Balastikova B, et al. Clinical and molecular-genetic markers of ADHD in children. Neuro Endocrinol Lett. 2008;29(3):320–7.
34. Alvarez-Arellano L, Gonzalez-Garcia N, Salazar-Garcia M, Corona JC. Antioxidants as a potential target against inflammation and oxidative stress in attention-deficit/hyperactivity disorder. Antioxidants. 2020;9(2):176.
35. Sciberras E, Mulraney M, Silva D, Coghill D. Prenatal risk factors and the etiology of ADHD-review of existing evidence. Curr Psychiatry Rep. 2017;19(1):1.
36. Park S, Cho SC, Kim JW, Shin MS, Yoo HJ, Oh SM, et al. Differential perinatal risk factors in children with attention-deficit/hyperactivity disorder by subtype. Psychiatry Res. 2014;219(3):609–16.
37. Khoshbakht Y, Bidaki R, Salehi-Abargouei A. Vitamin D status and attention deficit hyperactivity disorder: a systematic review and meta-analysis of observational studies. Adv Nutr. 2018;9(1):9–20.
38. Han JY, Kwon HJ, Ha M, Paik KC, Lim MH, Gyu Lee S, et al. The effects of prenatal exposure to alcohol and environmental tobacco smoke on risk for ADHD: a large population-based study. Psychiatry Res. 2015;225(1–2):164–8.
39. Sourander A, Sucksdorff M, Chudal R, Surcel HM, Hinkka-Yli-Salomaki S, Gyllenberg D, et al. Prenatal cotinine levels and ADHD among offspring. Pediatrics. 2019;143(3):e20183144.
40. Nigg JT, Breslau N. Prenatal smoking exposure, low birth weight, and disruptive behavior disorders. J Am Acad Child Adolesc Psychiatry. 2007;46(3):362–9.
41. Franz AP, Bolat GU, Bolat H, Matijasevich A, Santos IS, Silveira RC, et al. Attention-deficit/hyperactivity disorder and very preterm/very low birth weight: a meta-analysis. Pediatrics. 2018;141(1):e20171645.
42. Kim JH, Kim JY, Lee J, Jeong GH, Lee E, Lee S, et al. Environmental risk factors, protective factors, and peripheral biomarkers for ADHD: an umbrella review. Lancet Psychiatry. 2020;7(11):955–70.
43. Banerjee TD, Middleton F, Faraone SV. Environmental risk factors for attention-deficit hyperactivity disorder. Acta Paediatr. 2007;96(9):1269–74.
44. Banaschewski T, Becker K, Dopfner M, Holtmann M, Rosler M, Romanos M. Attention-deficit/hyperactivity disorder. Dtsch Arztebl Int. 2017;114(9):149–59.
45. Li L, Lagerberg T, Chang Z, Cortese S, Rosenqvist MA, Almqvist C, et al. Maternal pre-pregnancy overweight/obesity and the risk of attention-deficit/hyperactivity disorder in offspring: a systematic review, meta-analysis and quasi-experimental family-based study. Int J Epidemiol. 2020;49(3):857–75.
46. Chen Q, Sjolander A, Langstrom N, Rodriguez A, Serlachius E, D'Onofrio BM, et al. Maternal pre-pregnancy body mass index and offspring attention deficit hyperactivity disorder: a population-based cohort study using a sibling-comparison design. Int J Epidemiol. 2014;43(1):83–90.
47. Chang Z, Lichtenstein P, D'Onofrio BM, Almqvist C, Kuja-Halkola R, Sjolander A, et al. Maternal age at childbirth and risk for ADHD in offspring: a population-based cohort study. Int J Epidemiol. 2014;43(6):1815–24.
48. McCann D, Barrett A, Cooper A, Crumpler D, Dalen L, Grimshaw K, et al. Food additives and hyperactive behaviour in 3-year-old and 8/9-year-old children in the community: a randomised, double-blinded, placebo-controlled trial. Lancet. 2007;370(9598):1560–7.

49. Kennedy M, Kreppner J, Knights N, Kumsta R, Maughan B, Golm D, et al. Early severe institutional deprivation is associated with a persistent variant of adult attention-deficit/hyperactivity disorder: clinical presentation, developmental continuities and life circumstances in the English and Romanian Adoptees study. J Child Psychol Psychiatry Allied Discip. 2016;57(10):1113–25.
50. Biederman J. Attention-deficit/hyperactivity disorder: a selective overview. Biol Psychiatry. 2005;57(11):1215–20.
51. Gould KL, Coventry WL, Olson RK, Byrne B. Gene-environment interactions in ADHD: the roles of SES and chaos. J Abnorm Child Psychol. 2018;46(2):251–63.
52. Nigg J, Nikolas M, Burt SA. Measured gene-by-environment interaction in relation to attention-deficit/hyperactivity disorder. J Am Acad Child Adolesc Psychiatry. 2010;49(9):863–73.
53. Unsel Bolat G, Ercan ES, Salum GA, Bilac O, Massuti R, Uysal Ozaslan T, et al. Validity of proposed DSM-5 ADHD impulsivity symptoms in children. Eur Child Adolesc Psychiatry. 2016;25(10):1121–32.
54. Childress AC, Berry SA. Pharmacotherapy of attention-deficit hyperactivity disorder in adolescents. Drugs. 2012;72(3):309–25.
55. Shellenberg TP, Stoops WW, Lile JA, Rush CR. An update on the clinical pharmacology of methylphenidate: therapeutic efficacy, abuse potential and future considerations. Expert Rev Clin Pharmacol. 2020;13(8):825–33.
56. Faraone SV. The pharmacology of amphetamine and methylphenidate: relevance to the neurobiology of attention-deficit/hyperactivity disorder and other psychiatric comorbidities. Neurosci Biobehav Rev. 2018;87:255–70.
57. Briars L, Todd T. A review of pharmacological management of attention-deficit/hyperactivity disorder. J Pediat Pharmacol Therapeut. 2016;21(3):192–206.
58. Corona JC. Natural compounds for the management of Parkinson's disease and attention-deficit/hyperactivity disorder. Biomed Res Int. 2018;2018:4067597.
59. Clemow DB. Misuse of Methylphenidate. Curr Top Behav Neurosci. 2017;34:99–124.
60. Morton WA, Stockton GG. Methylphenidate abuse and psychiatric side effects. Primary Care Compan J Clin Psychiatry. 2000;2(5):159–64.
61. Leonard BE, McCartan D, White J, King DJ. Methylphenidate: a review of its neuropharmacological, neuropsychological and adverse clinical effects. Hum Psychopharmacol. 2004;19(3):151–80.
62. Koda K, Ago Y, Cong Y, Kita Y, Takuma K, Matsuda T. Effects of acute and chronic administration of atomoxetine and methylphenidate on extracellular levels of noradrenaline, dopamine and serotonin in the prefrontal cortex and striatum of mice. J Neurochem. 2010;114(1):259–70.
63. Reed VA, Buitelaar JK, Anand E, Day KA, Treuer T, Upadhyaya HP, et al. The safety of atomoxetine for the treatment of children and adolescents with attention-deficit/hyperactivity disorder: a comprehensive review of over a decade of research. CNS Drugs. 2016;30(7):603–28.
64. Mohammadi MR, Akhondzadeh S. Pharmacotherapy of attention-deficit/hyperactivity disorder: nonstimulant medication approaches. Expert Rev Neurother. 2007;7(2):195–201.
65. Swanson CJ, Perry KW, Koch-Krueger S, Katner J, Svensson KA, Bymaster FP. Effect of the attention deficit/hyperactivity disorder drug atomoxetine on extracellular concentrations of norepinephrine and dopamine in several brain regions of the rat. Neuropharmacology. 2006;50(6):755–60.
66. Bymaster FP, Katner JS, Nelson DL, Hemrick-Luecke SK, Threlkeld PG, Heiligenstein JH, et al. Atomoxetine increases extracellular levels of norepinephrine and dopamine in prefrontal cortex of rat: a potential mechanism for efficacy in attention deficit/hyperactivity disorder. Neuropsychopharmacology. 2002;27(5):699–711.
67. Giovannitti JA Jr, Thoms SM, Crawford JJ. Alpha-2 adrenergic receptor agonists: a review of current clinical applications. Anesth Prog. 2015;62(1):31–9.

68. Alamo C, Lopez-Munoz F, Sanchez-Garcia J. Mechanism of action of guanfacine: a postsynaptic differential approach to the treatment of attention deficit hyperactivity disorder (adhd). Actas Esp Psiquiatr. 2016;44(3):107–12.
69. Arnsten AF. The use of alpha-2A adrenergic agonists for the treatment of attention-deficit/hyperactivity disorder. Expert Rev Neurother. 2010;10(10):1595–605.
70. Connor DF, Rubin J. Guanfacine extended release in the treatment of attention deficit hyperactivity disorder in children and adolescents. Drugs Today. 2010;46(5):299–314.
71. Naguy A. Clonidine use in psychiatry: panacea or panache. Pharmacology. 2016;98(1–2):87–92.
72. Muthuraman M, Moliadze V, Boecher L, Siemann J, Freitag CM, Groppa S, et al. Multimodal alterations of directed connectivity profiles in patients with attention-deficit/hyperactivity disorders. Sci Rep. 2019;9(1):20028.
73. Cupertino RB, Soheili-Nezhad S, Grevet EH, Bandeira CE, Picon FA, Tavares MEA, et al. Reduced fronto-striatal volume in attention-deficit/hyperactivity disorder in two cohorts across the lifespan. Neuroimage Clin. 2020;28:102403.
74. Baroni A, Castellanos FX. Neuroanatomic and cognitive abnormalities in attention-deficit/hyperactivity disorder in the era of 'high definition' neuroimaging. Curr Opin Neurobiol. 2015;30:1–8.
75. Greven CU, Bralten J, Mennes M, O'Dwyer L, van Hulzen KJ, Rommelse N, et al. Developmentally stable whole-brain volume reductions and developmentally sensitive caudate and putamen volume alterations in those with attention-deficit/hyperactivity disorder and their unaffected siblings. JAMA Psychiatry. 2015;72(5):490–9.
76. Durston S, van Belle J, de Zeeuw P. Differentiating frontostriatal and fronto-cerebellar circuits in attention-deficit/hyperactivity disorder. Biol Psychiatry. 2011;69(12):1178–84.
77. Cortese S, Kelly C, Chabernaud C, Proal E, Di Martino A, Milham MP, et al. Toward systems neuroscience of ADHD: a meta-analysis of 55 fMRI studies. Am J Psychiatry. 2012;169(10):1038–55.
78. Cubillo A, Halari R, Smith A, Taylor E, Rubia K. A review of fronto-striatal and fronto-cortical brain abnormalities in children and adults with Attention Deficit Hyperactivity Disorder (ADHD) and new evidence for dysfunction in adults with ADHD during motivation and attention. Cortex. 2012;48(2):194–215.
79. Bailey T, Joyce A. The role of the thalamus in ADHD symptomatology and treatment. Appl Neuropsychol Child. 2015;4(2):89–96.
80. Davis AS, Pass LA, Finch WH, Dean RS, Woodcock RW. The canonical relationship between sensory-motor functioning and cognitive processing in children with attention-deficit/hyperactivity disorder. Arch Clin Neuropsychol. 2009;24(3):273–86.
81. Sathyanesan A, Zhou J, Scafidi J, Heck DH, Sillitoe RV, Gallo V. Emerging connections between cerebellar development, behaviour and complex brain disorders. Nat Rev Neurosci. 2019;20(5):298–313.
82. Genro JP, Kieling C, Rohde LA, Hutz MH. Attention-deficit/hyperactivity disorder and the dopaminergic hypotheses. Expert Rev Neurother. 2010;10(4):587–601.
83. Prince J. Catecholamine dysfunction in attention-deficit/hyperactivity disorder: an update. J Clin Psychopharmacol. 2008;28(3 Suppl 2):S39–45.
84. Klein MO, Battagello DS, Cardoso AR, Hauser DN, Bittencourt JC, Correa RG. Dopamine: functions, signaling, and association with neurological diseases. Cell Mol Neurobiol. 2019;39(1):31–59.
85. Del Campo N, Chamberlain SR, Sahakian BJ, Robbins TW. The roles of dopamine and noradrenaline in the pathophysiology and treatment of attention-deficit/hyperactivity disorder. Biol Psychiatry. 2011;69(12):e145–57.
86. Swanson JM, Kinsbourne M, Nigg J, Lanphear B, Stefanatos GA, Volkow N, et al. Etiologic subtypes of attention-deficit/hyperactivity disorder: brain imaging, molecular genetic and environmental factors and the dopamine hypothesis. Neuropsychol Rev. 2007;17(1):39–59.

87. Gold MS, Blum K, Oscar-Berman M, Braverman ER. Low dopamine function in attention deficit/hyperactivity disorder: should genotyping signify early diagnosis in children? Postgrad Med. 2014;126(1):153–77.
88. Napolitano A, Manini P, d'Ischia M. Oxidation chemistry of catecholamines and neuronal degeneration: an update. Curr Med Chem. 2011;18(12):1832–45.
89. Goldstein DS, Kopin IJ, Sharabi Y. Catecholamine autotoxicity. Implications for pharmacology and therapeutics of Parkinson disease and related disorders. Pharmacol Ther. 2014;144(3):268–82.
90. Neri M, Cerretani D, Fiaschi AI, Laghi PF, Lazzerini PE, Maffione AB, et al. Correlation between cardiac oxidative stress and myocardial pathology due to acute and chronic norepinephrine administration in rats. J Cell Mol Med. 2007;11(1):156–70.
91. Spencer WA, Jeyabalan J, Kichambre S, Gupta RC. Oxidatively generated DNA damage after Cu(II) catalysis of dopamine and related catecholamine neurotransmitters and neurotoxins: role of reactive oxygen species. Free Radic Biol Med. 2011;50(1):139–47.
92. Joseph N, Zhang-James Y, Perl A, Faraone SV. Oxidative stress and ADHD: a meta-analysis. J Atten Disord. 2015;19(11):915–24.
93. Lopresti AL. Oxidative and nitrosative stress in ADHD: possible causes and the potential of antioxidant-targeted therapies. Attent Deficit Hyperact Disord. 2015;7(4):237–47.
94. Corona JC. Role of oxidative stress and neuroinflammation in attention-deficit/hyperactivity disorder. Antioxidants. 2020;9(11):1039.
95. Cobley JN, Fiorello ML, Bailey DM. 13 reasons why the brain is susceptible to oxidative stress. Redox Biol. 2018;15:490–503.
96. Solleiro-Villavicencio H, Rivas-Arancibia S. Effect of chronic oxidative stress on neuroinflammatory response mediated by CD4(+)T cells in neurodegenerative diseases. Front Cell Neurosci. 2018;12:114.
97. Simpson DSA, Oliver PL. ROS generation in microglia: understanding oxidative stress and inflammation in neurodegenerative disease. Antioxidants. 2020;9(8):743.
98. Hanisch UK, Kettenmann H. Microglia: active sensor and versatile effector cells in the normal and pathologic brain. Nat Neurosci. 2007;10(11):1387–94.
99. de Araujo Boleti AP, de Oliveira Flores TM, Moreno SE, Anjos LD, Mortari MR, Migliolo L. Neuroinflammation: an overview of neurodegenerative and metabolic diseases and of biotechnological studies. Neurochem Int. 2020;136:104714.
100. Dunn GA, Nigg JT, Sullivan EL. Neuroinflammation as a risk factor for attention deficit hyperactivity disorder. Pharmacol Biochem Behav. 2019;182:22–34.
101. Lurie DI. An integrative approach to neuroinflammation in psychiatric disorders and neuropathic pain. J Exp Neurosci. 2018;12:1179069518793639.
102. Gilhus NE, Deuschl G. Neuroinflammation - a common thread in neurological disorders. Nat Rev Neurol. 2019;15(8):429–30.
103. Ni Chasaide C, Lynch MA. The role of the immune system in driving neuroinflammation. Brain Neurosci Adv. 2020;4:2398212819901082.
104. De Nardo D. Toll-like receptors: activation, signalling and transcriptional modulation. Cytokine. 2015;74(2):181–9.
105. Famitafreshi H, Karimian M. Prostaglandins as the agents that modulate the course of brain disorders. Degener Neurol Neuromuscul Dis. 2020;10:1–13.
106. Afridi R, Kim JH, Rahman MH, Suk K. Metabolic regulation of glial phenotypes: implications in neuron-glia interactions and neurological disorders. Front Cell Neurosci. 2020;14:20.
107. DiSabato DJ, Quan N, Godbout JP. Neuroinflammation: the devil is in the details. J Neurochem. 2016;139(Suppl 2):136–53.
108. Michael BD, Griffiths MJ, Granerod J, Brown D, Keir G, Wnek M, et al. The interleukin-1 balance during encephalitis is associated with clinical severity, blood-brain barrier permeability, neuroimaging changes, and disease outcome. J Infect Dis. 2016;213(10):1651–60.
109. Hawkins BT, Davis TP. The blood-brain barrier/neurovascular unit in health and disease. Pharmacol Rev. 2005;57(2):173–85.

110. Basu A, Krady JK, Enterline JR, Levison SW. Transforming growth factor beta1 prevents IL-1beta-induced microglial activation, whereas TNFalpha- and IL-6-stimulated activation are not antagonized. Glia. 2002;40(1):109–20.
111. Bazan NG. The docosanoid neuroprotectin D1 induces homeostatic regulation of neuroinflammation and cell survival. Prostaglandins Leukot Essent Fat Acids. 2013;88(1): 127–9.
112. Serhan CN. Pro-resolving lipid mediators are leads for resolution physiology. Nature. 2014;510(7503):92–101.
113. Tremblay ME, Stevens B, Sierra A, Wake H, Bessis A, Nimmerjahn A. The role of microglia in the healthy brain. J Neurosci. 2011;31(45):16064–9.
114. Prinz M, Jung S, Priller J. Microglia biology: one century of evolving concepts. Cell. 2019;179 (2):292–311.
115. Colonna M, Butovsky O. Microglia function in the central nervous system during health and neurodegeneration. Annu Rev Immunol. 2017;35:441–68.
116. Boche D, Perry VH, Nicoll JA. Review: activation patterns of microglia and their identification in the human brain. Neuropathol Appl Neurobiol. 2013;39(1):3–18.
117. Reus GZ, Fries GR, Stertz L, Badawy M, Passos IC, Barichello T, et al. The role of inflammation and microglial activation in the pathophysiology of psychiatric disorders. Neuroscience. 2015;300:141–54.
118. Liddelow SA, Barres BA. Reactive astrocytes: production, function, and therapeutic potential. Immunity. 2017;46(6):957–67.
119. Nosi D, Lana D, Giovannini MG, Delfino G, Zecchi-Orlandini S. Neuroinflammation: integrated nervous tissue response through intercellular interactions at the "whole system" scale. Cells. 2021;10(5):1195.
120. Philips T, Rothstein JD. Oligodendroglia: metabolic supporters of neurons. J Clin Invest. 2017;127(9):3271–80.
121. Zeis T, Enz L, Schaeren-Wiemers N. The immunomodulatory oligodendrocyte. Brain Res. 2016;1641(Pt A):139–48.
122. Tanabe S, Yamashita T. The role of immune cells in brain development and neurodevelopmental diseases. Int Immunol. 2018;30(10):437–44.
123. Tanabe S, Yamashita T. B lymphocytes: crucial contributors to brain development and neurological diseases. Neurosci Res. 2019;139:37–41.
124. Hirahara K, Nakayama T. CD4+ T-cell subsets in inflammatory diseases: beyond the Th1/Th2 paradigm. Int Immunol. 2016;28(4):163–71.
125. Filiano AJ, Gadani SP, Kipnis J. How and why do T cells and their derived cytokines affect the injured and healthy brain? Nat Rev Neurosci. 2017;18(6):375–84.
126. Filiano AJ, Xu Y, Tustison NJ, Marsh RL, Baker W, Smirnov I, et al. Unexpected role of interferon-gamma in regulating neuronal connectivity and social behaviour. Nature. 2016;535 (7612):425–9.
127. Quinnies KM, Cox KH, Rissman EF. Immune deficiency influences juvenile social behavior and maternal behavior. Behav Neurosci. 2015;129(3):331–8.
128. Conti P, Lauritano D, Caraffa A, Gallenga CE, Kritas SK, Ronconi G, et al. Microglia and mast cells generate proinflammatory cytokines in the brain and worsen inflammatory state: suppressor effect of IL-37. Eur J Pharmacol. 2020;875:173035.
129. Hendriksen E, van Bergeijk D, Oosting RS, Redegeld FA. Mast cells in neuroinflammation and brain disorders. Neurosci Biobehav Rev. 2017;79:119–33.
130. Ishii H, Yoshida M. [Inflammatory cytokines]. Nihon Rinsho. 2010;68(5):819–22.
131. Segman RH, Meltzer A, Gross-Tsur V, Kosov A, Frisch A, Inbar E, et al. Preferential transmission of interleukin-1 receptor antagonist alleles in attention deficit hyperactivity disorder. Mol Psychiatry. 2002;7(1):72–4.
132. Widera D, Mikenberg I, Elvers M, Kaltschmidt C, Kaltschmidt B. Tumor necrosis factor alpha triggers proliferation of adult neural stem cells via IKK/NF-kappaB signaling. BMC Neurosci. 2006;7:64.

133. Kaur N, Chugh H, Sakharkar MK, Dhawan U, Chidambaram SB, Chandra R. Neuroinflammation mechanisms and phytotherapeutic intervention: a systematic review. ACS Chem Neurosci. 2020;11(22):3707–31.
134. Alvarez-Arellano L, Díaz de León-Guerrero S, Meza-Sosa KF, Jiménez-Ferrer CI, Pérez-Martínez L. Neurodegenerative disorders and inflammation. J Chem Inf Model. 2013;
135. Montgomery SL, Bowers WJ. Tumor necrosis factor-alpha and the roles it plays in homeostatic and degenerative processes within the central nervous system. J NeuroImmune Pharmacol. 2012;7(1):42–59.
136. Choi SJ, Lee KH, Park HS, Kim SK, Koh CM, Park JY. Differential expression, shedding, cytokine regulation and function of TNFR1 and TNFR2 in human fetal astrocytes. Yonsei Med J. 2005;46(6):818–26.
137. Raffaele S, Lombardi M, Verderio C, Fumagalli M. TNF production and release from microglia via extracellular vesicles: impact on brain functions. Cells. 2020;9(10):2145.
138. Wajant H, Scheurich P. TNFR1-induced activation of the classical NF-kappaB pathway. FEBS J. 2011;278(6):862–76.
139. Wertz IE, Dixit VM. Ubiquitin-mediated regulation of TNFR1 signaling. Cytokine Growth Factor Rev. 2008;19(3–4):313–24.
140. Hsu MP, Frausto R, Rose-John S, Campbell IL. Analysis of IL-6/gp130 family receptor expression reveals that in contrast to astroglia, microglia lack the oncostatin M receptor and functional responses to oncostatin M. Glia. 2015;63(1):132–41.
141. West PK, Viengkhou B, Campbell IL, Hofer MJ. Microglia responses to interleukin-6 and type I interferons in neuroinflammatory disease. Glia. 2019;67(10):1821–41.
142. Marisa R, Reesha PR, Michal B. Cytokines in the CNS. Handb Exp Pharmacol. 2018;248:397.
143. Lobo-Silva D, Carriche GM, Castro AG, Roque S, Saraiva M. Balancing the immune response in the brain: IL-10 and its regulation. J Neuroinflammation. 2016;13(1):297.
144. Darwish AH, Elgohary TM, Nosair NA. Serum interleukin-6 level in children with attention-deficit hyperactivity disorder (ADHD). J Child Neurol. 2019;34(2):61–7.
145. Donev R, Thome J. Inflammation: good or bad for ADHD? Attent Deficit Hyperact Disord. 2010;2(4):257–66.
146. Hamed RA, Elmalt HA, Salama AAA, Hammouda SM, Youness ER, Abd-Allah NA, et al. MMP-2, MMP-9, TNF-α levels in relation to subtypes of attention deficit hyperactivity disorder. Biomed Pharmacol J. 2021;14:541–8.
147. Cortese S, Angriman M, Comencini E, Vincenzi B, Maffeis C. Association between inflammatory cytokines and ADHD symptoms in children and adolescents with obesity: a pilot study. Psychiatry Res. 2019;278:7–11.
148. Elsadek AE, Al-Shokary AH, Abdelghani WE, Kamal NM, Ibrahim AO, El-Shorbagy HH, et al. Serum levels of interleukin-6 and tumor necrosis factor alpha in children with attention-deficit hyperactivity disorder. J Pediatr Neurosci. 2020;15(4):402–8.
149. Chang JP, Mondelli V, Satyanarayanan SK, Chiang YJ, Chen HT, Su KP, et al. Cortisol, inflammatory biomarkers and neurotrophins in children and adolescents with attention deficit hyperactivity disorder (ADHD) in Taiwan. Brain Behav Immun. 2020;88:105.
150. Kozlowska A, Wojtacha P, Rowniak M, Kolenkiewicz M, Huang ACW. ADHD pathogenesis in the immune, endocrine and nervous systems of juvenile and maturating SHR and WKY rats. Psychopharmacology. 2019;236(10):2937–58.
151. Allred EN, Dammann O, Fichorova RN, Hooper SR, Hunter SJ, Joseph RM, et al. Systemic inflammation during the first postnatal month and the risk of attention deficit hyperactivity disorder characteristics among 10 year-old children born extremely preterm. J NeuroImmune Pharmacol. 2017;12(3):531–43.
152. O'Shea TM, Joseph RM, Kuban KC, Allred EN, Ware J, Coster T, et al. Elevated blood levels of inflammation-related proteins are associated with an attention problem at age 24 mo in extremely preterm infants. Pediatr Res. 2014;75(6):781–7.

153. Oades RD, Dauvermann MR, Schimmelmann BG, Schwarz MJ, Myint AM. Attention-deficit hyperactivity disorder (ADHD) and glial integrity: S100B, cytokines and kynurenine metabolism--effects of medication. Behav Brain Funct. 2010;6:29.
154. Donfrancesco R, Nativio P, Di Benedetto A, Villa MP, Andriola E, Melegari MG, et al. Anti-yo antibodies in children with ADHD: first results about serum cytokines. J Atten Disord. 2020;24(11):1497–502.
155. Smith TF, Anastopoulos AD, Garrett ME, Arias-Vasquez A, Franke B, Oades RD, et al. Angiogenic, neurotrophic, and inflammatory system SNPs moderate the association between birth weight and ADHD symptom severity. Am J Med Genet B Neuropsychiatr Genet. 2014;165B(8):691–704.
156. Ribases M, Hervas A, Ramos-Quiroga JA, Bosch R, Bielsa A, Gastaminza X, et al. Association study of 10 genes encoding neurotrophic factors and their receptors in adult and child attention-deficit/hyperactivity disorder. Biol Psychiatry. 2008;63(10):935–45.
157. Giana G, Romano E, Porfirio MC, D'Ambrosio R, Giovinazzo S, Troianiello M, et al. Detection of auto-antibodies to DAT in the serum: interactions with DAT genotype and psycho-stimulant therapy for ADHD. J Neuroimmunol. 2015;278:212–22.
158. Corominas-Roso M, Armario A, Palomar G, Corrales M, Carrasco J, Richarte V, et al. IL-6 and TNF-alpha in unmedicated adults with ADHD: relationship to cortisol awakening response. Psychoneuroendocrinology. 2017;79:67–73.
159. Monje ML, Toda H, Palmer TD. Inflammatory blockade restores adult hippocampal neurogenesis. Science. 2003;302(5651):1760–5.
160. Beroun A, Mitra S, Michaluk P, Pijet B, Stefaniuk M, Kaczmarek L. MMPs in learning and memory and neuropsychiatric disorders. Cell Mol Life Sci. 2019;76(16):3207–28.
161. Glessner JT, Li J, Wang D, March M, Lima L, Desai A, et al. Copy number variation meta-analysis reveals a novel duplication at 9p24 associated with multiple neurodevelopmental disorders. Genome Med. 2017;9(1):106.
162. Asherson P, Gurling H. Quantitative and molecular genetics of ADHD. Curr Top Behav Neurosci. 2012;9:239–72.
163. Rovira P, Demontis D, Sanchez-Mora C, Zayats T, Klein M, Mota NR, et al. Shared genetic background between children and adults with attention deficit/hyperactivity disorder. Neuropsychopharmacology. 2020;45(10):1617–26.
164. Toto M, Margari F, Simone M, Craig F, Petruzzelli MG, Tafuri S, et al. Antibasal ganglia antibodies and antistreptolysin O in noncomorbid ADHD. J Atten Disord. 2015;19(11):965–70.
165. Passarelli F, Donfrancesco R, Nativio P, Pascale E, Di Trani M, Patti AM, et al. Anti-Purkinje cell antibody as a biological marker in attention deficit/hyperactivity disorder: a pilot study. J Neuroimmunol. 2013;258(1–2):67–70.
166. Fan LW, Pang Y. Dysregulation of neurogenesis by neuroinflammation: key differences in neurodevelopmental and neurological disorders. Neural Regen Res. 2017;12(3):366–71.
167. Cherkasova MV, Hechtman L. Neuroimaging in attention-deficit hyperactivity disorder: beyond the frontostriatal circuitry. Can J Psychiatr. 2009;54(10):651–64.
168. Miyazaki C, Koyama M, Ota E, Swa T, Mlunde LB, Amiya RM, et al. Allergic diseases in children with attention deficit hyperactivity disorder: a systematic review and meta-analysis. BMC Psychiatry. 2017;17(1):120.
169. Mogensen N, Larsson H, Lundholm C, Almqvist C. Association between childhood asthma and ADHD symptoms in adolescence--a prospective population-based twin study. Allergy. 2011;66(9):1224–30.
170. Schans JV, Cicek R, de Vries TW, Hak E, Hoekstra PJ. Association of atopic diseases and attention-deficit/hyperactivity disorder: a systematic review and meta-analyses. Neurosci Biobehav Rev. 2017;74(Pt A):139–48.
171. Halfon N, Newacheck PW. Evolving notions of childhood chronic illness. JAMA. 2010;303(7):665–6.

172. Raison CL, Capuron L, Miller AH. Cytokines sing the blues: inflammation and the pathogenesis of depression. Trends Immunol. 2006;27(1):24–31.
173. Aguilar-Valles A, Inoue W, Rummel C, Luheshi GN. Obesity, adipokines and neuroinflammation. Neuropharmacology. 2015;96(Pt A):124–34.
174. Cortese S, Tessari L. Attention-deficit/hyperactivity disorder (ADHD) and obesity: update 2016. Curr Psychiatry Rep. 2017;19(1):4.
175. Albayrak O, Putter C, Volckmar AL, Cichon S, Hoffmann P, Nothen MM, et al. Common obesity risk alleles in childhood attention-deficit/hyperactivity disorder. Am J Med Genet B Neuropsychiatr Genet. 2013;162B(4):295–305.
176. Kang SS, Kurti A, Fair DA, Fryer JD. Dietary intervention rescues maternal obesity induced behavior deficits and neuroinflammation in offspring. J Neuroinflammation. 2014;11:156.
177. Ashwood P, Krakowiak P, Hertz-Picciotto I, Hansen R, Pessah I, Van de Water J. Elevated plasma cytokines in autism spectrum disorders provide evidence of immune dysfunction and are associated with impaired behavioral outcome. Brain Behav Immun. 2011;25(1):40–5.
178. Gumusoglu SB, Chilukuri ASS, Santillan DA, Santillan MK, Stevens HE. Neurodevelopmental outcomes of prenatal preeclampsia exposure. Trends Neurosci. 2020;43(4):253–68.
179. Novak CM, Ozen M, Burd I. Perinatal brain injury: mechanisms, prevention, and outcomes. Clin Perinatol. 2018;45(2):357–75.
180. Grizenko N, Shayan YR, Polotskaia A, Ter-Stepanian M, Joober R. Relation of maternal stress during pregnancy to symptom severity and response to treatment in children with ADHD. J Psychiatry Neurosci. 2008;33(1):10–6.
181. Grizenko N, Fortier ME, Gaudreau-Simard M, Jolicoeur C, Joober R. The effect of maternal stress during pregnancy on IQ and ADHD symptomatology. J Can Acad Child Adolesc Psychiatry. 2015;24(2):92–9.
182. Obernier JA, White AM, Swartzwelder HS, Crews FT. Cognitive deficits and CNS damage after a 4-day binge ethanol exposure in rats. Pharmacol Biochem Behav. 2002;72(3):521–32.
183. Terasaki LS, Schwarz JM. Effects of moderate prenatal alcohol exposure during early gestation in rats on inflammation across the maternal-fetal-immune interface and later-life immune function in the offspring. J NeuroImmune Pharmacol. 2016;11(4):680–92.
184. Huang L, Wang Y, Zhang L, Zheng Z, Zhu T, Qu Y, et al. Maternal smoking and attention-deficit/hyperactivity disorder in offspring: a meta-analysis. Pediatrics. 2018;141(1): e20172465.
185. Sengupta SM, Fortier ME, Thakur GA, Bhat V, Grizenko N, Joober R. Parental psychopathology in families of children with attention-deficit/hyperactivity disorder and exposed to maternal smoking during pregnancy. J Child Psychol Psychiatry Allied Discip. 2015;56(2): 122–9.
186. Zhu J, Zhang X, Xu Y, Spencer TJ, Biederman J, Bhide PG. Prenatal nicotine exposure mouse model showing hyperactivity, reduced cingulate cortex volume, reduced dopamine turnover, and responsiveness to oral methylphenidate treatment. J Neurosci. 2012;32(27):9410–8.
187. Herrmann M, King K, Weitzman M. Prenatal tobacco smoke and postnatal secondhand smoke exposure and child neurodevelopment. Curr Opin Pediatr. 2008;20(2):184–90.
188. Pineles BL, Park E, Samet JM. Systematic review and meta-analysis of miscarriage and maternal exposure to tobacco smoke during pregnancy. Am J Epidemiol. 2014;179(7): 807–23.
189. Hall HA, Speyer LG, Murray AL, Auyeung B. Prenatal maternal infections and children's neurodevelopment in the UK millennium cohort study: a focus on ASD and ADHD. J Atten Disord. 2022;26:616.
190. Ginsberg Y, D'Onofrio BM, Rickert ME, Class QA, Rosenqvist MA, Almqvist C, et al. Maternal infection requiring hospitalization during pregnancy and attention-deficit hyperactivity disorder in offspring: a quasi-experimental family-based study. J Child Psychol Psychiatry Allied Discip. 2019;60(2):160–8.

191. Rand KM, Austin NC, Inder TE, Bora S, Woodward LJ. Neonatal Infection and Later Neurodevelopmental Risk in the Very Preterm Infant. J Pediatr. 2016;170:97–104.
192. Ystrom E, Gustavson K, Brandlistuen RE, Knudsen GP, Magnus P, Susser E, et al. Prenatal exposure to acetaminophen and risk of ADHD. Pediatrics. 2017;140(5):e20163840.
193. Chen MH, Pan TL, Wang PW, Hsu JW, Huang KL, Su TP, et al. Prenatal exposure to acetaminophen and the risk of attention-deficit/hyperactivity disorder: a nationwide study in Taiwan. J Clin Psychiatry. 2019;80(5):18m12612.
194. Gustavson K, Ask H, Ystrom E, Stoltenberg C, Lipkin WI, Suren P, et al. Maternal fever during pregnancy and offspring attention deficit hyperactivity disorder. Sci Rep. 2019;9(1): 9519.
195. Gustafsson HC, Sullivan EL, Battison EAJ, Holton KF, Graham AM, Karalunas SL, et al. Evaluation of maternal inflammation as a marker of future offspring ADHD symptoms: a prospective investigation. Brain Behav Immun. 2020;89:350–6.
196. Han VX, Patel S, Jones HF, Dale RC. Maternal immune activation and neuroinflammation in human neurodevelopmental disorders. Nat Rev Neurol. 2021;17(9):564–79.
197. Tessari L, Angriman M, Diaz-Roman A, Zhang J, Conca A, Cortese S. Association between exposure to pesticides and ADHD or autism spectrum disorder: a systematic review of the literature. J Atten Disord. 2022;26:48.
198. Roberts JR, Karr CJ. Council on environmental H. Pesticide exposure in children. Pediatrics. 2012;130(6):e1765–88.
199. Rochester JR. Bisphenol A and human health: a review of the literature. Reprod Toxicol. 2013;42:132–55.
200. Hansen JB, Bilenberg N, Timmermann CAG, Jensen RC, Frederiksen H, Andersson AM, et al. Prenatal exposure to bisphenol A and autistic- and ADHD-related symptoms in children aged 2 and5 years from the Odense Child Cohort. Environ Health. 2021;20(1):24.
201. Minatoya M, Kishi R. A review of recent studies on bisphenol a and phthalate exposures and child neurodevelopment. Int J Environ Res Public Health. 2021;18(7):3585.
202. Choi G, Villanger GD, Drover SSM, Sakhi AK, Thomsen C, Nethery RC, et al. Prenatal phthalate exposures and executive function in preschool children. Environ Int. 2021;149: 106403.
203. Papalou O, Kandaraki EA, Papadakis G, Diamanti-Kandarakis E. Endocrine disrupting chemicals: an occult mediator of metabolic disease. Front Endocrinol. 2019;10:112.
204. Lenters V, Iszatt N, Forns J, Cechova E, Kocan A, Legler J, et al. Early-life exposure to persistent organic pollutants (OCPs, PBDEs, PCBs, PFASs) and attention-deficit/hyperactivity disorder: a multi-pollutant analysis of a Norwegian birth cohort. Environ Int. 2019;125:33–42.
205. Skogheim TS, Villanger GD, Weyde KVF, Engel SM, Suren P, Oie MG, et al. Prenatal exposure to perfluoroalkyl substances and associations with symptoms of attention-deficit/ hyperactivity disorder and cognitive functions in preschool children. Int J Hyg Environ Health. 2020;223(1):80–92.
206. Simoes LR, Sangiogo G, Tashiro MH, Generoso JS, Faller CJ, Dominguini D, et al. Maternal immune activation induced by lipopolysaccharide triggers immune response in pregnant mother and fetus, and induces behavioral impairment in adult rats. J Psychiatr Res. 2018;100:71–83.
207. Carias E, Hamilton J, Robison LS, Delis F, Eiden R, Quattrin T, et al. Chronic oral methylphenidate treatment increases microglial activation in rats. J Neural Transm (Vienna). 2018;125(12):1867–75.
208. Sadasivan S, Pond BB, Pani AK, Qu C, Jiao Y, Smeyne RJ. Methylphenidate exposure induces dopamine neuron loss and activation of microglia in the basal ganglia of mice. PLoS One. 2012;7(3):e33693.
209. Schmidt AJ, Krieg JC, Clement HW, Gebhardt S, Schulz E, Heiser P. Impact of drugs approved for treating ADHD on the cell survival and energy metabolism: an in-vitro study in human neuronal and immune cells. J Psychopharmacol. 2010;24(12):1829–33.

210. Motaghinejad M, Motevalian M, Shabab B, Fatima S. Effects of acute doses of methylphenidate on inflammation and oxidative stress in isolated hippocampus and cerebral cortex of adult rats. J Neural Transm (Vienna). 2017;124(1):121–31.
211. O'Sullivan JB, Ryan KM, Curtin NM, Harkin A, Connor TJ. Noradrenaline reuptake inhibitors limit neuroinflammation in rat cortex following a systemic inflammatory challenge: implications for depression and neurodegeneration. Int J Neuropsychopharmacol. 2009;12(5):687–99.
212. Park JH, Shin BN, Chen BH, Kim IH, Ahn JH, Cho JH, et al. Neuroprotection and reduced gliosis by atomoxetine pretreatment in a gerbil model of transient cerebral ischemia. J Neurol Sci. 2015;359(1–2):373–80.
213. Verlaet AAJ, Maasakkers CM, Hermans N, Savelkoul HFJ. Rationale for dietary antioxidant treatment of ADHD. Nutrients. 2018;10(4):405.
214. Moghadas M, Essa MM, Ba-Omar T, Al-Shehi A, Qoronfleh MW, Eltayeb EA, et al. Antioxidant therapies in attention deficit hyperactivity disorder. Front Biosci. 2019;24:313–33.
215. Alvarez-Arellano L, Salazar-Garcia M, Corona JC. Neuroprotective effects of quercetin in pediatric neurological diseases. Molecules. 2020;25(23):5597.
216. Richardson AJ. Omega-3 fatty acids in ADHD and related neurodevelopmental disorders. Int Rev Psychiatry. 2006;18(2):155–72.

# A Link Between Inflammatory Mechanisms and Fibromyalgia

16

Ashika Bains, Samuel Kohrman, Diana Punko, and Gregory Fricchione

**Abstract**

Fibromyalgia (FM) is a condition characterized by chronic widespread pain, which has traditionally been considered psychogenic in nature due to lack of known underlying organic pathophysiology. In more recent years, inflammation of the nervous system has become increasingly recognized as a sign of neuropsychiatric conditions, and this association may enhance our knowledge of conditions such as FM. Emerging evidence has suggested inflammation, particularly neuroinflammation, as a potential contributor underlying the etiology of FM. Studies have searched for linked biomarkers with mixed results, though the literature is beginning to point to increased systemic levels of pro-inflammatory cytokines such as IL-6 and IL-8 in patients with FM relative to healthy controls. A multicenter imaging study has also reported results suggestive of microglial activation related to the presence of FM. Given the consistency in

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neuroinflammatory effects implicated in "sickness behavior" characteristic of chronic systemic inflammatory conditions such as cancer or rheumatic diseases, therein springs the hypothesis for a connection between FM and neuroinflammation as discussed in this chapter.

**Keywords**

Fibromyalgia · Neuroinflammation · Neuropsychiatry · Chronic stress · Chronic pain · Central sensitivity syndromes (CSS) · Pain · Nociception

## 16.1 Introduction

### 16.1.1 History of Fibromyalgia

Fibromyalgia (FM) is a complex condition that overlaps the fields of internal medicine, rheumatology, psychiatry, and immunology. It falls into a category of syndromes that historically has lacked disease-defining biological markers, leading to the phenomena often being regarded as "psychogenic" rather than "organic" in etiology. Other examples of conditions in this category, sometimes called medically unexplained symptoms or functional somatic syndromes, include chronic fatigue syndrome (CFS) or myalgic encephalomyelitis (ME), irritable bowel syndrome (IBS), and temporomandibular joint pain. The term *fibromyalgia* is relatively new, first recognized by the American Medical Association in 1987, but in the past, the disorder has gone by other names including *chronic widespread pain* and *myofascial pain syndrome*. Over one hundred years ago, Sir William Gowers coined the term *fibrositis,* but this nomenclature is no longer in use as it became apparent that inflammation of connective tissues was not a common pathological finding [1]. The later term *fibromyalgia* better captured the concept of pain in the muscle fibers without the requirement for local inflammation, though, as this chapter will show, central inflammation may be involved.

In 1990, the American College of Rheumatology (ACR) presented their first set of diagnostic criteria for FM, primarily intended for consistency in research settings rather than clinical diagnosis. These criteria included generalized pain for at least 3 months, in at least three of four quadrants of the body, with pain elicited at a minimum of 11 out of 18 "tender points" [2]. Since then, the ACR has released updates of these criteria in 2010, 2011, and again in 2016, which will be described further below. FM was first included in the World Health Organization's International Statistical Classification of Diseases and Related Health Problems (ICD) in 1992 [3]. In contrast to the 3 months' duration of symptoms required by other definitions, in the most recent edition, ICD-10, the definition of FM can include "an acute, subacute, or chronic" painful state, although it also describes FM as "a chronic disorder" [3].

It is worth noting that FM is frequently comorbid with mental health disorders and that the condition is accordingly at risk for diagnostic overshadowing. In the most recent edition of the American Psychiatric Association's (APA) *Diagnostic and Statistical Manual of Mental Disorders*, Edition 5 (DSM-5), which was published in 2013, FM would be best categorized as a subset of somatic symptom disorder with *predominant pain* as a specifier [4]. Interestingly, the DSM-5 diagnosis of somatic symptom disorder no longer requires exclusion of a known general medical condition or the presence of symptoms in excess of what would be expected of a related general medical condition as did its DSM-IV predecessor, somatization disorder [5], perhaps reflecting a more nuanced understanding of these conditions.

### 16.1.2 Clinical Phenotype and Diagnostic Criteria for Fibromyalgia

FM is characterized by pain that is widespread in distribution and chronic in duration. There are frequently additional symptoms that accompany FM including fatigue, subjective cognitive impairment, sleep disturbance, depression, and anxiety. These additional symptoms are better reflected in the revised ACR diagnostic criteria released most recently in 2016, which includes a numerical score, the fibromyalgia severity (FS) scale, that is the sum of the Widespread Pain Index (WPI) and Symptom Severity Scale (SSS) score [6]. The WPI addresses the pain component of the diagnosis, and the SSS score incorporates the presence and severity of nonpain symptoms including fatigue, cognitive dysfunction, and headache (see Table 16.1 for details). To meet criteria for diagnosis under the most recent schema, a patient must have generalized pain, the presence of symptoms for at least 3 months, and a WPI $\geq 7$ and SSS score $\geq 5$ *or* WPI 4–6 and SSS score $\geq 9$. These revised criteria additionally eliminated the requirement for "tender points" (which were shown to be difficult to reliably identify in everyday clinical practice) and also accommodates other illnesses that could present with similar symptoms.

Efforts continue to establish an evidence-based diagnostic system for FM. The Addiction Clinical Trial Translations, Innovations, Opportunities, and Networks (ACTTION), a public-private partnership with the United States Food and Drug Administration (FDA) and the American Pain Society (APS), initiated the ACTTION-APS Pain Taxonomy (AAPT) for improved clinical usefulness and consistency across chronic pain disorders [7]. Using the AAPT framework, an international group of FM clinical and research experts proposed a new definition of FM; however, it requires further evaluation for feasibility, reliability, and validity before being put into widespread use [8].

### 16.1.3 Epidemiology, Demographics, and Prevalence

Estimates of the prevalence of FM range from 2% to up to 8% of the population depending on the criteria used [9, 10], and FM occurs more commonly in women than in men [9, 11]. FM is often comorbid with other so-called medically

**Table 16.1** American College of Rheumatology's criteria for fibromyalgia

| | | |
|---|---|---|
| ***Criteria*** | | |
| A patient satisfies modified 2016 fibromyalgia criteria if the following 3 conditions are met: | | |
| 1. Widespread pain index (WPI) ≥7 and symptom severity scale (SSS) score ≥5 OR WPI of 4–6 and SSS score ≥9 | | |
| 2. Generalized pain, defined as pain in at least 4 of 5 regions, must be present. Jaw, chest, and abdominal pain are not included in generalized pain definition | | |
| 3. Symptoms have been generally present for at least 3 months | | |
| A diagnosis of fibromyalgia is valid irrespective of other diagnoses. A diagnosis of fibromyalgia does not exclude the presence of other clinically important illnesses | | |
| ***Ascertainment*** | | |
| 1. **WPI**: In how many areas has the patient had pain over the last week? The WPI score will be between 0 and 19 | | |
| Left upper region (Region 1) | Right upper region (Region 2) | Axial region (Region 5) |
| Jaw, left[a] | Jaw, right[a] | Neck |
| Shoulder girdle, left | Shoulder girdle, right | Upper back |
| Upper arm, left | Upper arm, right | Lower back |
| Lower arm, left | Lower arm, right | Chest[a] |
| Left lower region (Region 3) | Right lower region (Region 4) | Abdomen[a] |
| Hip (buttock, trochanter), left | Hip (buttock, trochanter), right | |
| Upper leg, left | Upper leg, right | |
| Lower leg, left | Lower leg, right | |
| 2. **SSS score** | | Scale: |
| For each of the three symptoms below, indicate the level of severity over the past week using the scale to the right: | | 0 = No problem |
| (a) Fatigue | | 1 = Slight or mild problems, generally mild or intermittent |
| (b) Waking unrefreshed | | 2 = Moderate, considerable problems, often present and/or at a moderate level |
| (c) Cognitive symptoms | | 3 = Severe, pervasive, continuous, life-disturbing problems |
| The **SSS score** is the sum of the severity scores of the 3 symptoms (fatigue, waking unrefreshed, and cognitive symptoms) plus the number of the following symptoms that the patient has been bothered by that occurred during the previous 6 months: | | |
| 1. Headaches | | |
| 2. Pain or cramps in lower abdomen | | |
| 3. Depression | | |
| The final SSS score will be between 0 and 12 | | |
| The **fibromyalgia severity (FS) scale** is the sum of the WPI and SSS (between 0 and 31) | | |

Source: Adapted from Wolfe et al. [6]
[a] Not included in generalized pain definition

unexplained syndromes or functional somatic syndromes including CFS, IBS, interstitial cystitis, and others. To date, there have been no identified useful clinical biomarkers for the diagnosis of FM; however, there is preliminary evidence of a cytokine/chemokine signature that can help distinguish FM from systemic lupus erythematosus (SLE) and rheumatoid arthritis [12]. Purported risk factors for FM include: female sex, older/middle age, the presence of musculoskeletal disorders, high body mass index, cigarette smoking, and lower educational level and socioeconomic status [10, 13].

### 16.1.4 Differential Diagnosis

The signs and symptoms of FM overlap considerably with those of other conditions, and these should be considered when clinically formulating a differential diagnosis. Although the revised ACR diagnostic criteria for FM described above have increased diagnostic precision, there are currently no clinically relevant objective tests for FM, and thus, proper evaluation for FM requires a comprehensive assessment by a skilled clinician.

CFS was previously discussed in this text as being in FM's same syndromic category. Some authors (e.g., [13]) suggest that in fact the two are not distinct disorders but rather exist on a spectrum with pain as the prominent feature at one extreme (i.e., FM) and fatigue at the other end of the spectrum (i.e., CFS) as demonstrated in Fig. 16.1. Both pain and fatigue are essentially subjective phenomena.

Neurasthenia is a related condition that was popularized in the years after the American Civil War by the neurologist George Miller Beard and was thought to arise out of nervous system exhaustion. Headaches, fatigue, and impotence were the reported symptoms, and the stress of modern civilization was the proposed cause. Although many consider this concept to be outdated, neurasthenia is still listed as a diagnosis in the ICD-10 [3] and in the most recent edition of the Chinese Society of Psychiatry's Chinese Classification of Mental Disorders in 2001 [14]. IBS is another somatic symptom condition that is linked to the same pathophysiology as FM [15] and shares some overlap in symptoms (namely, pain or cramping in the lower abdomen) but is distinct in that the pain is relieved by defecation.


**Fig. 16.1** Fibromyalgia and chronic fatigue syndrome as part of a spectrum. *FM* fibromyalgia, *CFS* chronic fatigue syndrome

FM can be distinguished from systemic inflammatory rheumatic diseases (such as SLE) by the presence of skin rashes, vasculitis, and adenopathy in the latter [16]. Additionally, systemic inflammatory rheumatic diseases may have abnormal joint findings on radiographic imaging whereas these would not be characteristic of FM. Other potential culprits to consider when diagnosing FM include endocrinopathies such as hypothyroidism, which can cause profound fatigue, and medications such as cholesterol-lowering statins, which can cause muscle pain.

A controversial diagnosis, chronic Lyme disease is characterized by prolonged fatigue and pain following the acute resolution of a *Borrelia burgdorferi* infection. There are strong advocates for this theory though there is no clear evidence basis in support of it as a condition. Similarly, a subset of survivors of coronavirus disease 2019 (COVID-19), caused by the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), are reporting neuropsychiatric sequelae long after acute resolution of the illness. This condition is characterized by fatigue, anxiety, and "brain fog" or poor concentration [17] and has been given the name post-acute syndrome of COVID-19 (PASC) [18] or "long-haul COVID" [19]. Clearly, there is an overlap in symptomatology with FM, though of course FM existed as a concept long before the recent pandemic. The etiology of long-haul COVID is believed to be related at least in part to the repercussions of the intense cytokine storm that takes place at the beginning of acute COVID-19 illness and may be related to persistent autoantibodies in the central nervous system (CNS) [20].

Neuroinflammation offers a potential explanatory model and unifying underlying pathophysiology for these conditions, perhaps a final common pathway leading to altered pain processing.

### 16.1.5 Neuroinflammation and Psychiatric Illness

Neuroinflammation is addressed in detail elsewhere in this book so here we will review a few brief principles. Psychosocial stressors can produce neuroinflammation creating a vulnerability to chronic pain and fatigue, two cardinal symptoms of FM [21–23]. The inflammatory response is primarily mediated by activated microglial cells through the production and release of pro-inflammatory cytokines and chemokines in a process very similar to that which is found in response to threats in infectious disease. Michael Maes and Joseph Levine classically identified the strong association between depression and markers of inflammation in the periphery and in cerebrospinal fluid (CSF) [24, 25], and animal and human studies have demonstrated that immune challenges can induce depressive-like "sickness behavior" [23, 26, 27]. Neuroinflammation is known to be associated with several neurologic disorders such as multiple sclerosis and neurodegenerative disorders such as Alzheimer's disease [28]. It is interesting to note that depressive symptoms are frequently early manifestations of these diseases before their more distinguishing neurological symptoms emerge. In this chapter, we will expand on this work to explore the potential role of neuroinflammation in FM.

### 16.1.6 Additional Etiologic Factors in FM

While this chapter will focus primarily on biologic mechanisms, it is important to also appreciate psychological and social contributors for a full understanding of the illness [29]. The etiology for FM is multifactorial and includes the impact of genetic and biological influences, cognitive factors, and adverse life events [30]. An integrated perspective that acknowledges all these contributors is the best basis for clinical practice.

Genetic factors likely play a role in FM; to date, a few candidate genes with compelling evidence for their association with FM have been identified, TAAR1, ZNF77, and C11orf40, but their role in the etiology of FM is as yet unproven [7, 31]. History of adverse childhood experiences (ACEs) is more common among individuals with FM [13, 32] as is that of psychological distress, particularly sexual assault and physical abuse [33]. Relatedly, in a cross-sectional study, women with FM showed lower percentages of secure attachment style while showing higher avoidant attachment and increased anxious-ambivalent attachment relative to healthy female controls [34]. Alexithymia, a condition in which individuals have difficulty identifying and describing subjective feelings, has been associated with FM [35] perhaps due to symptom vigilance or misinterpretation of amplified physical sensations.

The degree of contribution to actual disease development of these proposed etiological factors has yet to be elucidated; it may be that the aforementioned considerations could be causative in FM or simply consequential. Nevertheless, these psychosocial factors are often prime targets for therapies as will be discussed below. Though clear and direct links have not yet been demonstrated, it would not be surprising if these additional etiologic factors were mediated by a common neuroinflammation pathophysiological mechanism.

## 16.2 A Link Between Inflammation and Illness

### 16.2.1 Overview of Inflammation

Inflammation is a response of the immune system using interactions between specific cell types and signaling molecules to protect and defend the body. Cell types include white blood cells (leukocytes), which penetrate tissue for phagocytosis and antigen presentation, and endothelial cells, which detect inflammatory stimuli and mediate leukocyte trafficking. Signaling molecules include locally acting molecules (e.g., nitric oxide), chemokines, and cytokines. There are different types of immune responses: innate immunity is a nonspecific response triggered by danger-associated molecular patterns (DAMPs) including pathogen-associated (PAMPs) and non-pathogen-associated (non-PAMPs); and adaptive immunity refers to specialized response to pathogens via T and B lymphocytes. During an inflammatory reaction, the endothelium becomes more permeable to leukocytes that are drawn toward a high concentration of cytokines near the stimulus/pathogen. Within the tissues,

leukocyte cells such as monocytes differentiate to macrophages for phagocytosis and secrete additional signaling molecules, thereby amplifying the inflammatory response from local to systemic. Inflammation has built in mechanisms that aim to control the initial stimulus for protection, as an excessive and prolonged inflammatory response can contribute to damage and disease.

Cytokines are signaling proteins that regulate inflammation either by exacerbation or by reduction; they can be classified as either pro- or anti-inflammatory. Pro-inflammatory cytokines include interleukin (IL)-1, IL-12, IL-18, tumor necrosis factor (TNF), interferon-gamma (IFN-γ), and granulocyte-macrophage colony stimulating factor. Anti-inflammatory cytokines include IL-4, IL-10, IL-13, interferon-alpha (IFN-α), and transforming growth factor-beta (TGF-β). Pro- and anti- are not rigid designations; disease pathology and effects of inflammatory molecules depend on location and context of disease. Chemokines are small chemoattractant cytokine proteins that help direct leukocyte migration and positioning by binding to endothelial cell surface receptors.

### 16.2.2 Neuroinflammation

Neuroinflammation differs from peripheral inflammation in that it concerns specialized cell types and specialized endothelium, the blood brain barrier (BBB). Specialized cell types include microglia and astrocytes. Microglial cells are the yolk-sac-derived resident macrophages in the central nervous system (CNS). In response to cytokines or other signaling molecules of acute inflammation, they transform from an inactive state to an activated phagocytic state and release pro-inflammatory mediators. Microglial activation can be divided into M1 or M2 activation. M1 activation is stimulated by IFN-γ and TNF to produce an aggressive immune response and release of cytokines. M2 activation is stimulated by IL-4 and has roles in wound healing and regulation of the macrophage response. Astrocytes are glial cells that release pro-inflammatory signaling molecules working in tandem with microglia.

The BBB and the choroid plexus (CP) form the interface between the peripheral and central immune systems. Cytokine levels are known to modulate the permeability of the BBB by altering the resistance of tight gap junctions of endothelial cells, thereby providing a connection between peripheral and central inflammation. Chemokines are involved in chemotaxis (cell movement in response to a stimulus) of astrocytes and microglia in response to inflammatory stimuli. The movement of leukocytes across the BBB is also regulated by chemokines, another connection between peripheral and central inflammation. The circumventricular organs (CVOs), located along the cerebral ventricular surface, lack endothelial tight junction barriers unlike other areas of the BBB and have fenestrated capillaries. These CVOs contribute to functions such as thermal homeostasis, energy balance, chemoreception of blood-derived substances, and neuroinflammation [36]. Saper et al. [37] proposed a mechanism by which circulating cytokines can enter the CVOs through the fenestrated capillaries and act on cells in the CNS such as microglia and astrocytes

[37]. In other studies, the neurosecretory region of CVOs, particularly the neurohypophysis (NH), has been associated with neuroglial plasticity [36]. Additionally, the CP provides a port of entry into the CNS for immune cells and contains several immune cells (such as CNS-specific T cells, macrophages, etc.); the plexus may be the site by which microglia access cerebrospinal fluid (CSF) [38]. Recent COVID-19 research has suggested that the CP is instrumental in the crossover of peripheral inflammation to neuroinflammation in COVID-19 neuropsychiatric illness [39].

### 16.2.3 Cytokines and FM

There is growing evidence establishing the relationship between neuroinflammation and FM and related disorders. The release of neuropeptides and the subsequent neuroinflammatory changes in the brain, the spinal cord, and the periphery are thought to contribute to the symptoms of FM.

Current evidence suggests that cytokines, and especially chemokines, may have a role in the pathogenesis of FM and may be correlated with disease symptomatology. Several studies have reported higher levels of IL-8 in serum and plasma of FM patients [40, 41]. IL-8 is stimulated by substance P (SP) and mediates sympathetic pain. SP and the SP structurally related hemokinin-1 (HK-1) and corticotropin-releasing hormone (CRH) levels were also shown to be significantly elevated in the blood of 84 female FM patients compared with that of healthy controls (HCs) in one study [21]. IL-17 was increased in FM, correlated with levels of TNF, and is positively correlated with pain and anxiety [42]. IL-6, a cytokine secreted by neurons, glial cells, and endothelial cells, is associated with hyperalgesia, fatigue, and sympathetic system activation and was also found to be elevated in plasma of FM patients [40]. Higher levels of TNF-α were also reported in FM as compared with controls in blood; TNF-α has been reported to promote degeneration of BBB and is associated with fatigue and anorexia [43]. A number of studies suggest that pro-inflammatory cytokines such as IL-1, IL–6, and IL-8 are elevated, and anti-inflammatory cytokines such as IL-4 and IL-10 are lower or normal in FM patients [21]. One study showed a significant association between both IL-8 and IL-6 and clinical severity scores in patients with FM suggesting a link to a clinically relevant mechanism in FM [21].

A study by Bäckryd et al. [44] provided an analysis of 92 inflammatory-related proteins in CSF and plasma and established evidence for both neuroinflammation and chronic systemic inflammation in FM patients [44]. Participants were excluded if there was an organic etiology for pain or if the participant met criteria for a mood or anxiety disorder based on the DSM-IV. The investigation included the CSF and plasma inflammatory profiles of 40 FM patients compared with CSF from 10 HCs and plasma from 46 blood donor controls. Assessment of neuroinflammation was provided through CSF analysis and chronic systemic inflammation through plasma. In plasma, elevated levels of 21 inflammatory substances in FM patients were reported as compared with those of HCs. Four proteins were found to overlap in both CSF and plasma analyses. Notably, the authors also reported, in the FM group,

elevated levels of CSF chemokine CX3CL1 (also known as fractalkine), which is associated with pain. In addition, previously reported findings of high systemic levels of pro-inflammatory cytokines IL-6 and IL-8 were replicated in this study.

Üçeyler et al. [45] performed a systematic review and meta-analysis of reported studies measuring cytokine levels in FM [45]. Overall, they reported most studies were not of high methodological quality. The better-quality studies showed elevated plasma IL-8 levels and elevated IL-6 serum levels in FM patients compared with those of controls. The following results were reproduced regardless of the methodology used: patients with FM have higher serum levels of IL-1RA, IL-6, and IL-8; patients with FM have higher plasma levels of IL-8. Higher levels of these cytokines in plasma and/or serum of patients with FM may be associated with pain as previously discussed given IL-8's association with SP. Whether the findings concerning cytokines are a risk factor or a consequence of the pathological process is an important area for further research.

Of special interest is the finding of cytokine IL-8 elevation, but not elevated IL-1β, in the CSF of FM patients as compared with HCs. This cytokine is co-localized with the translocator protein (TSPO) in glial cells, which implies that IL-8 is derived from glial cells within the CNS, which mediate neuroinflammation [21].

### 16.2.4 Neuroinflammation and FM

While the consequences of neuroinflammation in FM are not yet clear, evidence suggests that neuroinflammation does play a role. Albrecht et al. [46] conducted a multicenter study measuring brain glial activation in 31 FM patients and 27 HCs using positron emission tomography (PET) scans [46]. Glial activation can be studied in vivo using PET scanning with labeled proteins such as [$^{11}$C]PBR28, which is upregulated in microglia and astrocytes under inflammatory conditions. [$^{11}$C]PBR28 binds to the high affinity state of the TSPO. They utilized [$^{11}$C]PBR28 PET ligand to study microglia and astrocyte activation while also utilizing [$^{11}$C]-L-deprenyl-$D_2$ signal PET scan, which is more specific for astrocytes, in a sub-study. Results showed [$^{11}$C]PBR28 distribution volume was elevated in several brain regions including dorsolateral prefrontal cortex (dlPFC), dorsomedial prefrontal cortex (dmPFC), primary somatosensory cortex (S1), primary motor cortex (M1), posterior cingulate cortex (PCC), supplementary motor area (SMA), supramarginal gyrus (SMG), and superior parietal lobule (SPL) in FM patients as compared with HCs. There were no regions where standardized uptake values normalized by occipital cortex signal were higher in HCs when compared with FM. They also reported a positive association between protein PET signals in several regions and subjective ratings of fatigue. These results suggest correlation between neuroinflammation and FM. No statistical difference was found in [$^{11}$C]-L-deprenyl-$D_2$ PET scans suggesting astrocyte activation may not be a prominent factor in FM pathophysiology.

As mounting evidence suggests a connection between neuroinflammation and FM, consideration must be paid to negative outcomes of chronic neuroinflammation and its potential relationship with the persistent symptoms of FM. In chronic neuroinflammation, microglia and astrocytes can remain activated for extended periods of time releasing excess quantities of cytokines and neurotoxic molecules. This can affect various brain functions associated with clinical manifestations of FM.

Microglia are involved in synaptic plasticity. A growing body of evidence suggests astrocytes may have a function in maintenance and development of the synapse; astrocyte-mediated inflammation could have detrimental long-term effects. Neurogenesis (the differentiation of progenitor cells to neurons) is inhibited by pro-inflammatory cytokines such as IL-6, TNF-α, and IL-18; microglia release these cytokines. The TNF family of cytokines causes direct biological effects on neuronal survival and apoptosis. Astrocytes and microglia release inducible nitric oxide, which, when elevated, can cause neuronal apoptosis [47]. Microglia in the thalamus are associated with maintaining the pain sensation even after the original stimulus is no longer present, and therefore chronic microglial activation may mediate chronic pain.

## 16.3 Discussion

### 16.3.1 Stress and Its Relation to FM Symptoms

When bearing in mind effects of life-stress and its relation to FM symptoms, a potential link to consider is a nucleic acid-sensing pathway linking to the immune response. A protein named *cyclic-GMP-AMP synthase* (cGAS) is an innate immune system receptor, which detects pathogenic DNA and alerts an innate immune adaptor protein, *stimulator of interferon genes* or STING, to mount an IFN-based response to protect the host. Chronic low-grade inflammation can engage the cGAS-STING system as it senses cytosolic self-DNA in the form of mitochondrial DNA (mtDNA), which has slipped through mitochondrial pores disrupted by reactive oxygen species (ROS) [48]. Stress-induced non-PAMP inflammation alone can be associated with a neuroinflammatory cytokine profile related to gene expression upregulation of nuclear factor kB (NF- kB) and type I IFN pathways. This inflammation has been shown to be driven by the cGAS-STING pathway [49]. Excessive engagement of the cGAS–STING pathway in the brain (especially by microglia) can lead to neuroinflammation and neurodegeneration. Future targeting of the cGAS–STING pathway may afford therapeutic benefits in disorders such as FM [48].

### 16.3.2 Potential Model for Neuroinflammation and Symptoms

As discussed above, one theoretical connection between neuroinflammation and FM symptoms is the impact on selectively vulnerable brain regions with increased BBB permeability to chronic non-PAMPs inflammation.

These areas referred to as CVOs include regions such as the median eminence (ME), organum vasculosum of the lamina terminalis (OVLT), subfornical organ (SFO), area postrema (AP), neurohypophysis (NH), and the pineal gland. The neurons within these CVOs innervate the nearby portions of the brain; the ME and OVLT project to the hypothalamus, anterior cingulate cortex (ACC), and basal forebrain. The SFO projects to the hypothalamus. The pineal gland is susceptible to inflammation, and the AP abuts the brainstem and cerebellum [37]. As such, symptoms stemming from neuroinflammation across numerous disease states are believed to derive from a number of pathways impacted by these regions. These can be conceptualized as the aforementioned "sickness behaviors" including the core components of FM: depression, fatigue, impaired sleep, and cognitive dysfunction [23, 50]. This inflammatory milieu is also implicated in FM pain symptoms.

### 16.3.3 Pain Symptoms

Literature suggests that the pain component of FM and that of other central sensitivity syndromes (CSS) may be associated with nociceptive system hypersensitivity characterized by increased transmission, central amplification, reduced inhibitor control mechanisms, and reduced opponent non-nociceptive sensory processing [51]. The afferent and non-nociceptive pathways and their component regions are presumed to be interconnected and function in a circuit, leading dysfunction in one region to directly or indirectly impact other regions. Several ascending nociceptive afferent pathway regions include the thalamus, amygdala, posterior, mid and anterior insula, midbrain, and dorsal anterior cingulate cortex (dACC). Additional regions such as the secondary somatosensory regions, adjacent opercula, and inferior frontal gyrus [52] are connected via projections from the more directly impacted regions [53].

Furthermore, additional areas in the descending nociceptive inhibitory pathway such as the perigenual ACC (pgACC), the posterior cingulate (PCC), precuneus, and paracentral lobule have been associated with a variety of affective, autonomic, social, self-referential, and decision-making functions also related to FM symptomatology [52]. The triad of the ACC, insular cortex, and amygdala is theorized as the confluence of emotional pain and somatic pain. All play roles in propagating and mitigating both anxiety and chronic pain. Particularly, the ACC and its related network appear to play a special role in illness-related anxiety and anxiety related to somatic pain and suffering [54].

The connection between neuroinflammation and pain may be related to chemokines, specifically the CXL family, which are expressed by neurons and glial cells and initiate cytokine activations leading to neuroinflammation. As stated above, one of the proteins found to be significantly elevated in both CSF and plasma in individuals with FM is fractalkine (also known as CX3CL1), which is linked to the signaling pathway supposed to be most prominent in experimental models of neuropathic pain [55]. Fractalkine is released from primary afferent terminals by cathepsin S. Activated microglia release cathepsin S, which then cleaves fractalkine

from neurons. If fractalkine is increased in CSF and plasma in FM patients, it could mean an increase in cathepsin S and indicate microglia activation. Increased levels of cathepsin S and/or fractalkine may provide contributions to etiology of pain in FM patients.

From a neurotransmitter perspective, neuropathic pain is transmitted to the CNS via a number of receptors including serotonergic 5-hydroxytryptamine receptors (5-HT1B/D and 5-HT3), alpha 2 adrenergic receptors (α2), gamma aminobutyric acid (GABA) receptors, glutamate receptors, and *N*-methyl-D-aspartate (NMDA) receptors [56]. A 2016 review by Chinn et al. identified the involvement of 5-HT2A, alpha 1 adrenergic receptors (α1), and catechol-*o*-methyltransferase (COMT) pathways in FM symptoms [51]. In one theory, NMDA serves a long-term potentiation (LTP) role of pain in the ACC. Central sensitivity plays a role in symptoms, and lack of restorative slow wave non-rapid eye movement (N-REM) sleep is presumed to enhance pain sensitivity [51]. Oxidative stress is implicated, particularly leading to decreased levels of catalase and coenzyme Q10 (CoQ10); micronutrient deficiencies (vitamins D, B1, B12/folate) are also theorized to play a role [51, 57].

### 16.3.4 Mood and Behavioral Symptoms

In addition to pain, symptoms of fatigue, anxiety, depression, non-restorative sleep, and cognitive impairment predominate in FM. Neuroinflammatory and oxidative disruptions at analogous regions to the ME and OVLT project to the adjacent ACC and basal forebrain leading to aberrations in pathways of orexin, histamine, acetylcholine (Ach), GABA, and glutamate; disruptions in these pathways along with pathways of monoamines 5-HT, dopamine (DA), NE are associated with dysregulated mood, behavior, sleep, and cognition [58, 59]. Additionally, TNF-α, IL-1, and IL-6 disrupt the LTP of the hippocampus disrupting memory, as studied in postoperative inflammatory states [23, 60].

The OVLT abuts the ACC. From the oncology literature, inflammation allows INF-α to increase regional blood flow in the dACC. This increased dACC activity has been demonstrated in individuals at risk for mood and anxiety disorders [23].

The ME and OVLT both project toward the hypothalamus. The SFO projects to the hypothalamus with GABA projections from the pallidothalamic circuit. Neuroinflammation is known to increase the neuropeptide CRH leading to flattened diurnal cortisol release with increased secretion at times of traditional quiescence, decreased glucocorticoid sensitivity, and decreased cortisol responsiveness, all of which are found in conditions like depression, insomnia, anxiety, and anorexia [23].

The AP abuts the brainstem and cerebellum and impacts the projection path of DA release in the ventral tegmental area (VTA), norepinephrine (NE) release in the locus coeruleus (LC), and serotonin or 5-HT release in the raphe nuclei (RN) with impact on mood and behavior as mentioned above. As such, inflammation at the AP leads to CNS serotonin depletion, particularly at the RN. IFN and IL-6 deplete tryptophan via induction of indolamine 2,3 dioxygenase (IDO), also reducing production of melatonin with a shift to production of neurotoxic kynurenine

[58, 61]. Furthermore, TNF-α and lL-1 increase the function of the 5-HT and NE pumps, increasing their reuptake and decreasing these monoamines in their respective clefts, influencing mood and behavior [23].

CNS dopaminergic neurons in the basal ganglia (BG) and VTA are known to be susceptible to neuroinflammation and oxidative stress. Namely, as studied in Parkinson's disease, neuroinflammation causes DA neuronal cell damage with involvement of IL-6, TNF, and IL-1β [62]. Neuroinflammation-driven TNF triggers proteolytic activation of protein kinase Cδ (PKCδ), leading to proapoptotic signaling and dopaminergic cell death, while tumor necrosis factor-R1 (TNF-R1) neutralize antibodies and the soluble TNF receptor etanercept blocked TNF-induced PKCδ proteolytic activation [63]. Oxidative stress is also involved in the pathology of Parkinson's disease (PD), destabilizing DA neurons and oxidizing DA molecules to dopamine quinone, which in turn can propagate further ROS [62].

The pineal gland appears relatively susceptible to oxidative stress and hypoxic damage [64]. Melatonin synthesis is modulated by GABA and glutamate while release is regulated by direct NE signals [59, 65], all of which is dysregulated in association with neuroinflammation and can lead to downstream sleep-wake cycle dysregulation [58].

## 16.4 Treatment Considerations

### 16.4.1 Nonpharmacologic Treatment

Treatment for FM is multimodal and includes lifestyle, pharmacologic, and somatic treatments. These treatments are tailored to relieve symptoms attributed to the proposed mechanisms above. While first-line treatment remains education and a physical exercise plan, more comprehensive multimodal treatment is now considered the gold standard, without a consensus as to which components should be included. The FIBROWALK study [66] demonstrated that a multicomponent treatment plan including neuroscience education, therapeutic exercise, cognitive behavioral therapy (CBT), and mindfulness in addition to pharmacologic treatment as usual had more favorable revised FM Impact Questionnaire (FIQR) scores than pharmacologic treatment as usual alone in terms of physical impairment, pain, kinesiophobia, and physical function more so than in fatigue, anxiety, and depressive symptoms.

As the aforementioned descending inhibitory nociceptive pathway is moderated by cognitive biases such as negative maladaptive thoughts and emotional/behavioral factors, which can lead to a perceived experience of pain [52, 66], education and psychotherapy permitting reconceptualization are presumed to mitigate this [51, 53]. CBT has shown to strengthen self-efficacy and promote adaptive coping strategies in patients suffering from chronic pain; a meta-analysis of 29 randomized controlled trials (RCTs) testing the effectiveness of CBT-based interventions for FM noted significant and small to medium mean effect sizes in pain relief, improvement of quality of life, reduction of negative mood, disability, and fatigue [66].

Exercise, namely aerobic exercise, has shown benefit in domains of pain, with mixed results in fatigue, given that the exercise itself could be fatiguing. Mind-body therapy, namely yoga, Tai-Chi, and Qigong have shown benefit in domains of pain and sleep, with unclear outcomes for other domains [51]. Exercise has further shown decrease in pain sensitivity and significantly greater activity in the anterior insula and the left dlPFC [53].

Acupuncture has been studied with nociceptive pain benefit and is directly related to the aforementioned afferent and non-afferent nociceptive tracts. In a review by Ong et al. [53], nociceptive pain benefit from acupuncture was correlated with increased connectivity between insula and mid cingulate cortex. Furthermore, the benefit was correlated with greater activation in the dlPFC, ACC, and midbrain as shown by PET study, while functional magnetic resonance imaging (fMRI) study showed greater activation in the amygdala, ACC, periaqueductal gray (PAG), hypothalamus, anterior insula, and PFC [53]. In rat and mouse models, acupuncture serves a potential anti-inflammatory role, particularly in relation to dopaminergic cells [67].

### 16.4.2 Pharmacologic Treatment

Despite the potential etiology of FM being inflammatory, traditional anti-inflammatory therapeutics including nonsteroidal anti-inflammatory drugs (NSAIDs) and glucocorticoids do not seem to be effective for pain in FM [68]. These treatments target the cyclooxygenase pathway, whereas it is suspected that the inflammatory downstream aspects of FM are more related to sympathetic overarousal and decreased responsiveness of monocytes.

Pharmacologically, tricyclic antidepressants (TCAs), namely amitriptyline, have been studied as the first-line agent for FM. The mechanism of action includes serotonin and NE reuptake inhibition and direct effects on 5-HT receptors, muscarinic/anticholinergic receptors, histamine receptor antagonism, α1 receptor antagonism, voltage-gated sodium channels, and some theoretical activity on opiate and NMDA/glutamate receptors [59]. Typical doses of 10–50 mg nightly improve pain, fatigue, sleep disturbance in FM in a superior fashion to that of selective serotonin reuptake inhibitors (SSRIs) and serotonin norepinephrine reuptake inhibitors (SNRIs), while the effect on depression requires more study as a monotherapy [51]. Amitriptyline does have evidence as antidepressant monotherapy throughout the psychiatric literature though usually at higher doses. In terms of FM, amitriptyline performed better in combination with SSRI fluoxetine than amitriptyline monotherapy [51]. This could theoretically be due to fluoxetine's role as a cytochrome P450 (CYP450) 2D6 inhibitor, a hepatic enzyme that is responsible for degradation of TCAs, thus increasing relative amitriptyline concentrations.

Some have suggested cyclobenzaprine, as included in Table 16.2, as an alternative initial agent given the structural similarity to amitriptyline. A single systematic review showed benefit at standard dosing (10–40 mg at night) for sleep but not pain [69] while another RCT of very low dose cyclobenzaprine (1–4 mg at night) showed

**Table 16.2** Pharmacologic agents in the treatment of FM

| Agent | Pharmacologic targets | Symptom domain | | | |
|---|---|---|---|---|---|
| | | Pain | Fatigue | Sleep | Depression |
| Amitriptyline[a, b] | 5-HT; NE/α-1/α-2; Ach; H1; NMDA/Glut | + | + | + | + |
| Cyclobenzaprine[c, d] | Undetermined; 5-HT2 antagonist at brainstem | +/− | − | +/− | − |
| SNRI[a] | 5-HT; NE/α-1/α-2 | + | − | − | + |
| SSRI[a] | 5-HT | +/− | +/− | +/− | + |
| Gabapentinoids[a] (pregabalin/ gabapentin) | α2δ voltage gated Ca channel; NMDA/Glut | + | − | + | − |
| Cannabinoids[a] | Indirect NMDA/glutamate | + | − | + | − |
| Sodium oxybate[a] | GABA | − | + | + | − |
| Low-dose naltrexone[e] | Opiate partial agonist | + | − | − | − |
| Memantine[f] | NMDA/glutamate | + | − | − | + |
| CoQ10[g] | Antioxidant | + | − | − | + |

In this table, + indicates that the given agent has beneficial effect in this symptom domain, − indicates that the given agent showed no benefit in this symptom domain, and +/− indicates that the evidence is mixed

[a] Chinn et al. [51]
[b] Lawson [59]
[c] Macfarlane et al. [69]
[d] Moldofsky et al. [70]
[e] Patten et al. [71]
[f] Olivan-Blázquez et al. [72]
[g] Lowry et al. [57]

self-reported benefit for both pain and sleep and for fatigue [70]. In general, it does not confer notable antidepressant benefit.

Second-line pharmacotherapy includes SNRIs, particularly duloxetine and milnacipran, both of which are approved by the United States FDA for FM. Duloxetine at a target dose of 60 mg daily confers pain and depression benefit, with no effect on fatigue and inconclusive effect on sleep as characterized by the FIQR and the mental component of quality of life (QoL) measures [51].

SSRIs such as fluoxetine showed benefit at higher than standard dosing (80 mg), and others such as citalopram, escitalopram, sertraline, and paroxetine have also been studied [51].

Pregabalin (FDA-approved for FM) along with gabapentin can act by binding and blocking the α2δ subunit of voltage-gated calcium ion channels to block over-excitation of neurotransmitters such as glutamate to reduce anxiety, worry, and fear, while also acting on such channels to block transmission of noxious mechanical and sub-noxious thermal stimuli from the peripheral nervous system (PNS) to the CNS [56]. They have demonstrated benefit for pain and sleep, with unclear effect on fatigue [51].

Some evidence exists for agents such as sodium oxybate, a salt derived from GABA, which binds GABA receptors and increases slow wave non-REM sleep, improving sleep disturbance, fatigue, and overall functioning [51].

Cannabinoids, which indirectly act as anti-glutaminergic agents, have also resulted in improved pain and FIQR scores in some studies while impacting only sleep in others [51].

### 16.4.3 Additional Agents

Additional agents with less robust evidence and exciting theoretical benefit, with low-cost low-harm implementation include:

Low dose naltrexone at a typical goal dose 4.5 mg has shown benefit in domains of pain and overall QoL in a number of small RCTs in patients with FM (e.g., [71]).

Memantine, an NMDA antagonist, showed improvement in self-reported pain and depression at doses of 20 mg daily in one small study [72]. In one model, LTP of pain and pain-related anxiety in the ACC is triggered by NMDA receptors. Increased phosphorylation of NMDA receptors in spinal dorsal horns theoretically increases central sensitization [57]. These effects may explain the benefit of anti-glutamatergic and NMDA antagonist agents in the treatment of FM such as amitriptyline, gabapentin/pregabalin, cannabinoids, and memantine.

Melatonin, suggested in 2020 as an anti-inflammatory and antioxidant agent in COVID-19 by Zhang et al., offers interesting conceptual benefit as an adjuvant medication in the regulation of immune system, inflammation, and oxidative stress in FM [73]. It theoretically acts on the sirtuin-1 (SIRT-1) pathway downregulating polarization of macrophages, decreasing their pro-inflammatory activation and subsequent release of pro-inflammatory cytokines/ILs. Melatonin theoretically downregulates (NF-kB), which is closely associated with inflammatory and oxidative response. Melatonin stimulates NF-E2-related factor 2 (Nrf2), which also confers theoretical benefit. As a result, melatonin downregulates TNF-α, IL-1β, IL-6, and IL-8 and elevates IL-10, an anti-inflammatory cytokine. Melatonin may upregulate anti-oxidative enzyme superoxide dismutase, downregulate pro-oxidative enzyme nitric oxide synthetase, and interact directly as a free radical scavenger [73]. Melatonin itself confers antioxidant and free radical scavenger effects; its absence would perpetuate further oxidative and pro-inflammatory states [64, 73].

Dietary interventions, such as nutritional changes or supplementation can aid patients in terms of reducing oxidative damage, modulate inflammatory states, improve energy production, and aid neuromodulation within the PNS and CNS [57]. A 2020 systematic review by Lowry et al., with relatively smaller sample sizes and lack of methodological homogeneity, identified the following:

– A diet low in fermentable oligo-di-mono-saccharides and polyols (FODMAP) appeared to improve nearly all related symptoms including pain, fatigue, sleep,

depression, and memory. This may be due to proposed antioxidant and neuromodulatory benefits.
- Supplementation with CoQ10 at doses 300–400 mg daily led to improvement in reported pain and depression symptoms. CoQ10 is decreased in oxidative stress and inflammatory conditions such as FM, depression, and chronic fatigue. This mitochondrial electron transport carrier molecule improves mitochondrial function and also serves as an antioxidant.
- Vitamin D at 50,000 IU once weekly did not separate from placebo, while vitamin D 1200–2400 IU daily did separate from placebo, with reported benefit in pain and morning fatigue. This may be explained by anti-inflammatory and neuromodulatory benefits.
- A combination of Vitamin C and Vitamin E alone did not separate from placebo, while the addition of *Nigella sativa* seeds did separate in terms of pain improvement. The proposed mechanism is by antioxidant effect, and vitamin C serves as an important cofactor in monoamine synthesis.

## 16.5 Conclusion

This chapter highlights the potential role of neuroinflammatory biomarkers in FM leading to a deeper understanding of this condition as opposed to the oversimplified "psychogenic" explanation. Importantly, specific therapeutic targets can be identified serving as possible niches for novel treatments for this often refractory condition. For instance, modulating microglial activity may be a possible therapeutic niche in fibromyalgia and potentially other centrally mediated pain syndromes [22]. From an evolutionary standpoint, a so-called trigger-happy inflammation response to stressors was advantageous for our ancestors; however, as our environment has changed to one with chronic psychosocial stress and noncommunicable diseases, this sensitive neuroinflammatory response has become increasingly maladaptive [27]. Although here we focus on FM, it is important to note somatization is a feature of other psychiatric conditions including depression; thus, this topic has broader implications. In conclusion, recent advances in knowledge of potential biomarkers of FM can help us elucidate neurobiology and evolutionary implications, which can promote change in perspective on the etiology of this and other central pain syndromes.

## References

1. Mbuyi N. Fibromyalgia. In: Ferri FF, editor. Ferri's clinical advisor 2022. Amsterdam: Elsevier; 2022. p. 627–629.e1.
2. Wolfe F, Smythe HA, Yunus MB, Bennett RM, Bombardier C, Goldenberg DL, et al. The American college of rheumatology 1990 criteria for the classification of fibromyalgia. Arthritis Rheum. 1990;33(2):160–72.

3. World Health Organization and WHO Collaborating Centers for Classification of Diseases. International statistical classification of diseases and related health problems (ICD-10). 10th revision. Geneva: World Health Organization; 1992.
4. American Psychiatric Association. Diagnostic and statistical manual of mental disorders: DSM-5. 5th ed. Arlington, VA: American Psychiatric Association; 2013.
5. American Psychiatric Association. Diagnostic and statistical manual of mental disorders: DSM-IV. 4th ed. Washington, DC: American Psychiatric Association; 1994.
6. Wolfe F, Clauw DJ, Fitzcharles M-A, Goldenberg DL, Häuser W, Katz RL, et al. 2016 revisions to the 2010/2011 fibromyalgia diagnostic criteria. Semin Arthritis Rheum. 2016;46(3):319–29.
7. Fillingim RB, Bruehl S, Dworkin RH, Dworkin SF, Loeser JD, Turk DC, et al. The ACTTION-American Pain Society Pain Taxonomy (AAPT): an evidence-based and multidimensional approach to classifying chronic pain conditions. J Pain. 2014;15(3):241–9.
8. Arnold LM, Bennett RM, Crofford LJ, Dean LE, Clauw DJ, Goldenberg DL, et al. AAPT diagnostic criteria for fibromyalgia. J Pain. 2019;20(6):611–28.
9. Jones GT, Atzeni F, Beasley M, Flüß E, Sarzi-Puttini P, Macfarlane GJ. The prevalence of fibromyalgia in the general population: a comparison of the American College of Rheumatology 1990, 2010, and modified 2010 classification criteria. Arthritis Rheumatol. 2015;67(2):568–75.
10. Creed F. A review of the incidence and risk factors for fibromyalgia and chronic widespread pain in population-based studies. Pain. 2020;161(6):1169–76.
11. Branco JC, Bannwarth B, Failde I, Abello Carbonell J, Blotman F, Spaeth M, et al. Prevalence of fibromyalgia: a survey in five European countries. Semin Arthritis Rheum. 2010;39(6): 448–53.
12. Wallace DJ, Gavin IM, Karpenko O, Barkhordar F, Gillis BS. Cytokine and chemokine profiles in fibromyalgia, rheumatoid arthritis and systemic lupus erythematosus: a potentially useful tool in differential diagnosis. Rheumatol Int. 2015;35(6):991–6.
13. Sharpe MC, O'Malley PG. Chapter 25: Chronic fatigue and fibromyalgia syndromes. In: Levenson J, editor. The American Psychiatric Association publishing textbook of psychosomatic medicine and consultation-liaison psychiatry. 3rd ed. Washington, DC: American Psychiatric Association Publishing; 2019. p. 709–36.
14. Chinese Society of Psychiatry. Chinese classification of mental disorders (CCMD-3). 3rd ed. Chinese Society of Psychiatry; 2001.
15. Feng B, La JH, Schwartz ES, Gebhart GF. Irritable bowel syndrome: methods, mechanisms, and pathophysiology. Neural and neuro-immune mechanisms of visceral hypersensitivity in irritable bowel syndrome. Am J Physiol Gastrointest Liver Physiol. 2012;302(10):G1085–98.
16. Häuser W, Perrot S, Sommer C, Shir Y, Fitzcharles M-A. Diagnostic confounders of chronic widespread pain: not always fibromyalgia. Pain Rep. 2017;2(3):e598.
17. Huang C, Huang L, Wang Y, Li X, Ren L, Gu X, et al. 6-month consequences of COVID-19 in patients discharged from hospital: a cohort study. Lancet. 2021;397(10270):220–32.
18. Nalbandian A, Sehgal K, Gupta A, Madhavan MV, McGroder C, Stevens JS, et al. Post-acute COVID-19 syndrome. Nat Med. 2021;27(4):601–15.
19. Rubin R. As their numbers grow, COVID-19 "long haulers" stump experts. JAMA. 2020;324 (14):1381.
20. Franke C, Ferse C, Kreye J, Reincke SM, Sanchez-Sendin E, Rocco A, et al. High frequency of cerebrospinal fluid autoantibodies in COVID-19 patients with neurological symptoms. Brain Behav Immun. 2021;93:415–9.
21. Littlejohn G, Guymer E. Neurogenic inflammation in fibromyalgia. Semin Immunopathol. 2018;40(3):291–300.
22. Duque L, Fricchione G. Fibromyalgia and its new lessons for neuropsychiatry. Med Sci Monitor Basic Res. 2019;25:169–78.
23. Miller AH, Ancoli-Israel S, Bower JE, Capuron L, Irwin MR. Neuroendocrine-immune mechanisms of behavioral comorbidities in patients with cancer. J Clin Oncol. 2008;26:971–82.

24. Maes M, Meltzer HY, Bosmans E, Bergmans R, Vandoolaeghe E, Ranjan R, et al. Increased plasma concentrations of interleukin-6, soluble interleukin-6, soluble interleukin-2 and transferrin receptor in major depression. J Affect Disord. 1995;34(4):301–9.
25. Levine J, Barak Y, Chengappa KNR, Rapoport A, Rebey M, Barak V. Cerebrospinal cytokine levels in patients with acute depression. Neuropsychobiology. 1999;40(4):171–6.
26. Raison CL, Capuron L, Miller AH. Cytokines sing the blues: inflammation and the pathogenesis of depression. Trends Immunol. 2006;27(1):24–31.
27. Miller AH, Raison CL. The role of inflammation in depression: from evolutionary imperative to modern treatment target. Nat Rev Immunol. 2016;16(1):22–34.
28. Heneka MT, Carson MJ, El Khoury J, Landreth GE, Brosseron F, Feinstein DL, et al. Neuroinflammation in Alzheimer's disease. Lancet Neurol. 2015;14(4):388–405.
29. Engel GL. The need for a new medical model: a challenge for biomedicine. Science (New York, NY). 1977;196(4286):129–36. https://pubmed.ncbi.nlm.nih.gov/847460/.
30. Adams L, Turk D. Psychosocial factors and central sensitivity syndromes. Curr Rheumatol Rev. 2015;11(2):96–108.
31. Feng J, Zhang Z, Wu X, Mao A, Chang F, Deng X, et al. Discovery of potential new gene variants and inflammatory cytokine associations with fibromyalgia syndrome by whole exome sequencing. PLoS One. 2013;8(6):e65033.
32. Coppens E, van Wambeke P, Morlion B, Weltens N, Giao Ly H, Tack J, et al. Prevalence and impact of childhood adversities and post-traumatic stress disorder in women with fibromyalgia and chronic widespread pain. Eur J Pain. 2017;21(9):1582–90.
33. Haviland MG, Morton KR, Oda K, Fraser GE. Traumatic experiences, major life stressors, and self-reporting a physician-given fibromyalgia diagnosis. Psychiatry Res. 2010;177(3):335–41.
34. Peñacoba C, Perez-Calvo S, Blanco S, Sanroman L. Attachment styles, pain intensity and emotional variables in women with fibromyalgia. Scand J Caring Sci. 2018;32(2):535–44.
35. di Tella M, Castelli L. Alexithymia and fibromyalgia: clinical evidence. Front Psychol. 2013;4:909.
36. Miyata S. New aspects in fenestrated capillary and tissue dynamics in the sensory circumventricular organs of adult brains. Front Neurosci. 2015;9:390.
37. Saper C, Breder C. The neurologic basis of fever. N Engl J Med. 1994;330(26):1880–6.
38. Pollak TA, Drndarski S, Stone JM, David AS, McGuire P, Abbott NJ. The blood–brain barrier in psychosis. Lancet Psychiatry. 2018;5:79–92.
39. Yang AC, Kern F, Losada PM, Agam MR, Maat CA, Schmartz GP, et al. Dysregulation of brain and choroid plexus cell types in severe COVID-19. Nature. 2021;595(7868):565–71.
40. Rodriguez-Pintó I, Agmon-Levin N, Howard A, Shoenfeld Y. Fibromyalgia and cytokines. Immunol Lett. 2014;161(2):200–3.
41. Kadetoff D, Lampa J, Westman M, Andersson M, Kosek E. Evidence of central inflammation in fibromyalgia—increased cerebrospinal fluid interleukin-8 levels. J Neuroimmunol. 2012;242(1–2):33–8.
42. Theoharides TC, Tsilioni I, Bawazeer M. Mast cells, neuroinflammation and pain in fibromyalgia syndrome. Front Cell Neurosci. 2019;13:353.
43. Bazzichi L, Rossi A, Massimetti G, Giannaccini G, Giuliano T, de Feo F, et al. Cytokine patterns in fibromyalgia and their correlation with clinical manifestations. Clin Exp Rheumatol. 2007;25(2):225.
44. Bäckryd E, Tanum L, Lind AL, Larsson A, Gordh T. Evidence of both systemic inflammation and neuroinflammation in fibromyalgia patients, as assessed by a multiplex protein panel applied to the cerebrospinal fluid and to plasma. J Pain Res. 2017;10:515–25.
45. Üçeyler N, Häuser W, Sommer C. Systematic review with meta-analysis: cytokines in fibromyalgia syndrome. BMC Musculoskel Disord. 2011;12:245.
46. Albrecht DS, Forsberg A, Sandström A, Bergan C, Kadetoff D, Protsenko E, et al. Brain glial activation in fibromyalgia—a multi-site positron emission tomography investigation. Brain Behav Immun. 2019;75:72–83.

47. Lyman M, Lloyd D, Ji X, Vizcaychipi M, Ma D. Neuroinflammation: the role and consequences. Neurosci Res. 2014;79:1–2.
48. Paul BD, Snyder SH, Bohr VA. Signaling by cGAS–STING in neurodegeneration, Neuroinflammation, and aging. Trends Neurosci. 2021;44:83–96.
49. Barrett TJ, Corr EM, van Solingen C, Schlamp F, Brown EJ, Koelwyn GJ, et al. Chronic stress primes innate immune responses in mice and humans. Cell Rep. 2021;36(10):109595.
50. Erickson MA, Banks WA. Neuroimmune axes of the blood–brain barriers and blood–brain interfaces: bases for physiological regulation, disease states, and pharmacological interventions. Pharmacol Rev. 2018;70(2):278–314.
51. Chinn S, Caldwell W, Gritsenko K. Fibromyalgia pathogenesis and treatment options update. Curr Pain Headache Rep. 2016;20:25.
52. López-Solà M, Woo CW, Pujol J, Deus J, Harrison BJ, Monfort J, et al. Towards a neurophysiological signature for fibromyalgia. Pain. 2017;158(1):34–47.
53. Ong WY, Stohler CS, Herr DR. Role of the prefrontal cortex in pain processing. Mol Neurobiol. 2019;56:1137–66.
54. Zhuo M. Neural mechanisms underlying anxiety-chronic pain interactions. Trends Neurosci. 2016;39:136–45.
55. Ryabkova VA, Churilov LP, Shoenfeld Y. Neuroimmunology: what role for autoimmunity, neuroinflammation, and small fiber neuropathy in fibromyalgia, chronic fatigue syndrome, and adverse events after human papillomavirus vaccination? Int J Mol Sci. 2019;20(20):5164.
56. Stahl SM. Stahl's essential psychopharmacology: neuroscientific basis and practical applications. 4th ed. New York, NY: Cambridge University Press; 2013. p. 608, xv, 608–xv
57. Lowry E, Marley J, McVeigh JG, McSorley E, Allsopp P, Kerr D. Dietary interventions in the management of fibromyalgia: a systematic review and best-evidence synthesis. Nutrients. 2020;12:2664.
58. Maldonado JR. Neuropathogenesis of delirium: review of current etiologic theories and common pathways. Am J Geriatr Psychiatr. 2013;21(12):1190–222.
59. Lawson K. A brief review of the pharmacology of amitriptyline and clinical outcomes in treating fibromyalgia. Biomedicines. 2017;5(2):24.
60. Terrando N, Monaco C, Ma D, Foxwell BMJ, Feldmannc M, Maze M. Tumor necrosis factor-a triggers a cytokine cascade yielding postoperative cognitive decline. Proc Natl Acad Sci U S A. 2010;107(47):20518–22.
61. Couch Y, Martin CJ, Howarth C, Raley J, Khrapitchev AA, Stratford M, et al. Systemic inflammation alters central 5-HT function as determined by pharmacological MRI. NeuroImage. 2013;75:177–86.
62. He J, Zhu G, Wang G, Zhang F. Oxidative stress and neuroinflammation potentiate each other to promote progression of dopamine neurodegeneration. Oxid Med Cell Longev. 2020;2020:6137521.
63. Gordon R, Anantharam V, Kanthasamy AG, Kanthasamy A. Proteolytic activation of proapoptotic kinase protein kinase Cδ by tumor necrosis factor α death receptor signaling in dopaminergic neurons during neuroinflammation. J Neuroinflammation. 2012;9(1):82.
64. Reiter RJ. Oxidative damage in the central nervous system: protection by melatonin. Prog Neurobiol. 1998;56(3):359–84.
65. Aulinas A. Physiology of the pineal gland and melatonin. In: Feingold KR, Anawalt B, Boyce A, Chrousos G, de Herder WW, Dhatariya K, et al., editors. Endotext. South Dartmouth, MA: MDText.com; 2000. http://www.ncbi.nlm.nih.gov/books/NBK550972/.
66. Serrat M, Sanabria-Mazo JP, Almirall M, Musté M, Feliu-Soler A, Méndez-Ulrich JL, et al. Effectiveness of a multicomponent treatment based on pain neuroscience education, therapeutic exercise, cognitive behavioral therapy, and mindfulness in patients with fibromyalgia (FIBROWALK study): a randomized controlled trial. Phys Ther. 2021;101:pzab200.
67. Jang JH, Yeom MJ, Ahn S, Oh JY, Ji S, Kim TH, et al. Acupuncture inhibits neuroinflammation and gut microbial dysbiosis in a mouse model of Parkinson's disease. Brain Behav Immun. 2020;89:641–55.

68. Derry S, Wiffen PJ, Häuser W, Mücke M, Tölle TR, Bell RF, et al. Oral nonsteroidal anti-inflammatory drugs for fibromyalgia in adults. Cochrane Database Syst Rev. 2017;2020(2).
69. Macfarlane GJ, Kronisch C, Dean LE, Atzeni F, Häuser W, Flub E, et al. EULAR revised recommendations for the management of fibromyalgia. Ann Rheum Dis. 2017;76(2):318–28.
70. Moldofsky H, Harris HW, Archambault WT, Kwong T, Lederman S. Effects of bedtime very low dose cyclobenzaprine on symptoms and sleep physiology in patients with fibromyalgia syndrome: a double-blind randomized placebo-controlled study. J Rheumatol. 2011;38(12): 2653–63.
71. Patten DK, Schultz BG, Berlau DJ. The safety and efficacy of low-dose naltrexone in the management of chronic pain and inflammation in multiple sclerosis, fibromyalgia, Crohn's disease, and other chronic pain disorders. Pharmacotherapy. 2018;38(3):382–9.
72. Olivan-Blázquez B, Herrera-Mercadal P, Puebla-Guedea M, Pérez-Yus M-C, Andrés E, Fayed N, et al. Efficacy of memantine in the treatment of fibromyalgia: a double-blind, randomised, controlled trial with 6-month follow-up. Pain. 2014;155(12):2517–25.
73. Zhang R, Wang X, Ni L, Di X, Ma B, Niu S, et al. COVID-19: melatonin as a potential adjuvant treatment. Life Sci. 2020;250:117583.

# Suicide and Inflammation

17

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**Abstract**

Suicide is a leading cause of death worldwide. Although the neurobiological dysfunction underlying suicidal behavior remains unclear, recent work suggests that the immune system may play a role in the pathophysiology of suicide. In this chapter, we discuss a nascent body of literature suggesting that peripheral and central nervous systems (CNS) inflammation are associated with suicidal behavior. Because early-life stress is a major risk factor for suicidal behavior and is also associated with immune dysregulation, we hypothesize that such immune dysregulation may be the mechanism by which childhood maltreatment leads to an increased risk of suicidal behavior and suicide. Targeting inflammatory processes may be a novel treatment strategy, especially in populations that have experienced childhood trauma and exhibit elevated inflammation. Future work should directly test the hypothesis that reducing inflammation would result in a reduction in suicidal behavior.

**Keywords**

Suicide · Inflammation · IL-6 · IL-1β · Microglia · Early-life adversity · TNF-α · Astrocyte · Kynurenine pathway

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## 17.1 Suicide

Suicide is a leading cause of death worldwide, with over 700,000 suicides occurring annually [1]. In the United States, suicide has become the tenth leading cause of death, and alarmingly the second leading cause of death in people ages 10–34 [2]. Each of these deaths impacts the lives of surviving friends, family, and communities, and over half of Americans report having been affected by a suicide [2]. Further, suicidal behavior places a significant psychological, social, and economic burden on society. For example, the average cost of one suicide is estimated at $1.4 million dollars, with the majority, 97%, of this cost due to lost productivity [3]. Suicide represents a significant, yet preventable public health concern; therefore, increased attention and funding have been devoted to suicide research and public awareness and prevention campaigns.

Suicidal phenotypes exist along a continuum, ranging from ideation to attempt to suicide, suggesting that this public health problem is not limited to those that die by suicide. In 2019 alone, nearly 1.4 million Americans made a suicide attempt, which can be defined as self-injury with the intention of death [2]. The cost of emergency room visits and hospitalizations associated with suicide attempts among young adults has been estimated at $2.6 billion annually [4]. Many others report suicidal ideation or having thoughts of ending their own life. Although estimates vary, a World Mental Health Survey found that the lifetime prevalence of suicidal ideation is 9.2% [5]. In the United States alone, 12 million Americans had serious thoughts of suicide in 2019 [2]. However, this number may be underestimated as a variety of factors can contribute to a person's hesitancy to report suicidal thoughts, including stigma, religious beliefs, and even criminalization of suicide in some countries. The frequency of suicidal ideation is positively correlated with the risk of attempting suicide, and a history of suicide attempts is a strong predictor of suicide death, with up to 40% of people that die by suicide having made a previous suicide attempt [6, 7].

Suicidal behavior is a global phenomenon; however, incidence varies between groups and across regions. Low- and middle-income countries, for example, account for nearly 80% of suicide deaths [1]. In addition, some regions, including Africa, Europe, and South-East Asia, tend to have higher suicide rates than the global average [1]. The lowest rates of suicide are found in the Eastern Mediterranean region [1]. Suicide rates also differ across age groups. In the United States in 2019, 36.6% of people that died by suicide were over 55 years old, compared to the 14.7% of suicides that occurred in 10–24 year olds [2]. Females are generally more likely to attempt suicide, while males are more likely to die by suicide [2]. Further, marginalized populations, including refugees, indigenous people, and prisoners tend to be at higher risk of suicidal behavior [5].

The vast majority, up to 90%, of suicides are associated with a diagnosable psychiatric illness [2, 8]. The most common psychiatric disorders associated with suicide include major depressive disorder (MDD), bipolar disorder (BD), substance use disorders, and schizophrenia [8]. The majority of patients diagnosed with these disorders will not experience suicidal behavior; however, certain features associated

with the specific disorder may predict suicide risk. For example, in MDD, the number, duration, and intensity of depressive episodes can influence the likelihood of suicidal behavior [9, 10]. In schizophrenia, patients with less insight into their illness may be at a greater risk for suicidal behavior [11]. Further, certain personality traits have been associated with an increased risk of suicidal behavior, including impulsivity and aggression, as well as perfectionism [12].

In addition to our understanding of the psychological risk factors for suicide, considerable effort has been made to determine the biological mechanisms that underlie suicidal behavior. The identification of biomarkers for suicide risk may lead to prevention strategies and is therefore an area of great research interest. Recently, psychiatric disorders associated with suicide, including MDD and schizophrenia, have been associated with increased inflammatory signaling, which has led to an increase in research into immune mechanisms underlying suicidal behavior.

## 17.2 The Immune System

The immune system was developed to protect mammals from invading microorganisms. This system involves a complex interplay between specialized cells and proteins that act to recognize foreign pathogens, contain the infection, and provide a memory in order to enhance future responses to the same pathogen. Importantly, multiple mechanisms have evolved to regulate the duration and magnitude of the immune response itself, which can cause damage to cells and tissues [13]. Further, the immune system can also act to repair tissue injuries that occur in the absence of infection. The immune system is made up of two main branches, the innate and adaptive immune systems, which work together to limit damage. The innate immune system serves as the first line of defense, providing rapid, nonspecific responses against invading pathogens. Cells of the innate immune system, including those of a myeloid lineage (e.g., macrophages/monocytes and dendritic cells) and lymphoid cells (e.g. natural killer cells), continuously monitor the circulation for conserved features of invading pathogens. Specialized receptors on the cell surface recognize damage-associated molecular patterns (DAMPs), host molecules that signal cellular damage, or pathogen-associated molecular patterns (PAMPs), which are located on infectious agents. Upon activation, cells of the innate immune system release cytokines, small proteins that act through receptors to influence cellular functions and migration. Cytokines can be either pro- or anti-inflammatory and have been divided into families based on their specialized roles. Among the pro-inflammatory cytokines, interleukin (IL)-6, tumor necrosis factor (TNF)-α, IL-1β, and the interferons (IFN) have been the most commonly studied in the psychoneuroimmunology field, while IL-10 is the most highly examined anti-inflammatory cytokine. The release of these cytokines can be influenced by not only a current infection but also genetics and previous immune challenges [14]. Cytokines are produced primarily by immune cells, but other cell types, including neurons and astrocytes of the central nervous system (CNS), can also produce and release cytokines.

In addition to the cellular components of the innate immune system, the complement system consists of ~20 soluble proteins that opsonize pathogens and induce inflammatory responses [15]. The complement system can be initiated along three distinct pathways: the classical pathway, the lectin pathway, and the alternative pathway. Each of these pathways ultimately leads to the cleavage of the inactive C3 protein into C3a, an inflammatory mediator, and C3b, an opsinin. During infection, complement activation and C3 cleavage ultimately results in inflammation and destruction of the invading pathogen [15].

The adaptive immune system, which produces a slower but more specific immune response, is made up of lymphocytes (T and B cells). Each lymphocyte, expressing a unique antigen receptor, exists in an inactive state until is activated by antigen recognition. Upon activation, lymphocytes undergo clonal differentiation to produce a specific immune response. T cells are made in the bone marrow but mature in the thymus. Upon activation, T cells can become one of three types of effector T cells. Cytotoxic T cells (CD8+ cells) detect infected cells and destroy them. T helper (Th) cells can influence the activity of other immune cells. Regulatory T cells (Treg) suppress the activity of other lymphocytes to prevent autoimmunity [13].

B cells, which are made and mature in the bone marrow, also express antigen-specific receptors. B cells are activated by Th cells that express the same receptor, leading to clonal proliferation and differentiation into plasma cells. These plasma cells then produce and release antibodies into the blood. Antibodies are Y-shaped proteins that recognize a unique part of the pathogen, called an antigen. Upon antigen binding, antibodies can act to neutralize microorganisms or target them for phagocytosis. A proportion of activated B and T cells become memory cells once the pathogen is removed from the body. These memory cells allow for a more rapid and robust immune response when the same antigen is encountered in the future [13].

## 17.3 Peripheral Versus Central Immunity

The brain was once considered an immune privileged organ, based on the observation that bacteria, viruses, vectors, and tissue grafts can evade immune recognition when delivered directly to the brain parenchyma [16]. However, more recent data suggest that immune privilege may not be absolute, as multiple brain compartments, including the ventricles, blood brain barrier (BBB), and choroid plexus actually contain immune cells, such as macrophages and dendritic cells. In addition, factors such as age or prior infection have been shown to increase baseline immune activity in the brain [16]. Importantly, the brain also contains specialized macrophages, called microglia, that colonize the central nervous system during early development and make up ~5–10% of all cells in the adult brain. Under healthy conditions, microglia exist in a highly ramified state, with fine, motile processes that continuously survey the microenvironment [17]. These "resting" microglia have been shown to play an active role in processes such as neurogenesis, neuronal differentiation, synaptic formation, pruning, and plasticity [18]. In addition to these homeostatic mechanisms, microglia can also respond to a wide variety of stimuli, including

infection or injury, by altering their morphology [18]. Microglia exist on a continuum of morphological phenotypes ranging from ramified to the highly activated amoeboid phenotype, which participate in antigen presentation, release cytokines, and remove cellular debris by phagocytosis [19].

Although not considered immune cells per say, astrocytes can also participate in central immune function. Under healthy conditions, astrocytes regulate important processes, including ion homeostasis, neurotransmitter reuptake, and synaptic plasticity [20]. Similar to microglia, astrocytes express receptors that detect a variety of stimuli, including those that signal infection or injury, and can respond by entering a range of activation states. Reactive astrocytes have been shown to release immune signaling molecules, including cytokines and complement proteins [21], and can be toxic to neurons and other brain cells. In addition to their role in immune signaling, astrocytes also contribute to the BBB, which helps to exclude foreign pathogens and immune cells from the brain. The BBB is composed of multiple cellular elements, including the end feet of astrocytic processes that surround blood vessels and provide a barrier between the brain and the circulation [22].

In addition to immune signaling that originates within the brain itself, multiple pathways have been shown to transduce immune signals from the periphery to the central nervous system [23]. One way in which peripheral immune signaling can activate a central immune response is via circumventricular organs, structures that surround brain ventricles and lack a functional BBB. Circumventricular organs, such as the median eminence, typically regulate autonomic nervous system activity and endocrine function; therefore, blood vessels in these organs contain fenestrations to allow diffusion of molecules through the blood vessel wall [24]. Cytokines can enter the brain parenchyma at these sites via volume diffusion [25]. In addition, in situations of high peripheral inflammation, cytokine transporters at the BBB can also move cytokines into the brain. Further, activation of IL-1 receptors on perivascular macrophages and endothelial cells of brain venules can result in the production of inflammatory mediators, such as prostaglandin E2 [26, 27]. Together, these pathways can transduce peripheral inflammation into central inflammation in the absence of the invasion of immune cells into the brain.

## 17.4 Evidence for Role of Immune Dysregulation in Suicide

### 17.4.1 Suicide Associated with Inflammatory Treatments

The hypothesis that inflammation may lead to suicidal behavior was first proposed when clinicians observed that immunotherapy was associated with depression and suicidal behavior in some patients. Recombinant cytokines, including IL-2, and INF-α, have been used to treat certain types of cancer and chronic viral infections, such as hepatitis C [28–30]. IFN-α has both antiviral and antiproliferative actions, but treatment produces significant side effects, including symptoms consistent with major depression in up to 50% of patients [31]. Further, multiple case reports presented in the 1980s and 1990s described patients, often with no psychiatric

history, that developed psychiatric side effects during immunotherapy, including suicidal ideation [32, 33]. A subset of these case studies also report suicide attempts and completions [33–36]. In some of the cases, symptoms improved upon the completion of cytokine therapy [32, 34–36]; however, others attempted suicide after the withdrawal of cytokine therapy [36]. These case studies suggest that there may be a connection between immunotherapy and suicidal behavior, and a limited number of subsequent experiments have confirmed this connection. One study examined the psychological consequences of IL-2 or IFN-α therapy on patients being treated for renal cell carcinoma or advanced melanoma [37]. Each form of immunotherapy examined, including IL-2 therapy alone, IFN-α therapy alone, and combined IL-2 + IFN-α treatment produced a significant increase in the severity of depressive episode as measured by the Montgomery-Asberg Depression Rating Scale (MADRS). The overall increase in MADRS score was caused, in part, by an increase in cognitive symptoms, which included suicidal thoughts [37]. Another study found that veterans treated with IFN-α for hepatitis C experienced a significant increase in suicidal thoughts during the course of cytokine therapy [38]. These results suggest that activation of the immune system may lead to an increased risk for depression and suicidal behavior.

### 17.4.2 Suicide Associated with Inflammatory Disorders

In addition to results suggesting that cytokine therapy may induce symptoms of depression and suicidal behavior, there is also evidence that a variety of medical diagnoses that involve activation of the immune system are also associated with an increased incidence of suicide. Autoimmune disorders, for example, occur when the body cannot distinguish between self and foreign invaders, leading to the attack of healthy cells, tissues, and organs by the immune system. Approximately 80 different autoimmune disorders that targeting different parts of the body have been identified. In multiple sclerosis (MS), the immune system attacks the cells that insulate nerves and facilitate electrical signaling. Patients with multiple sclerosis have an increased risk of dying by suicide. One epidemiological study found that MS patients were twice as likely as the general population to die by suicide [39]. However, other reports have found that MS patients are 7.5 times more likely than the general population to die by suicide [40]. Suicide risk seems to be increased even in autoimmune disorders that don't target the central nervous system. Psoriasis is another autoimmune disease in which T cells of the immune system attack skin cells. Patients with psoriasis have an increased risk of suicidal ideation compared to both healthy controls and those with other dermatological conditions (melanoma and allergy) [41–44]. Interestingly, blocking IL-17 signaling with an anti-IL-17RA increases suicidality in psoriasis patients [45], whereas targeting pathogenic Th17 cells with IL-17A therapy improved depression symptoms in this population [46].

Further, infections have also been associated with an increased risk of suicide. One Danish population study found that people who were hospitalized for infection were more likely to die by suicide than people that were not hospitalized. Further,

this relationship was graded, with increasing number of infections associated with an increased risk of suicide [47]. This effect has been observed across pathogen types, including bacterial, viral, and parasitic infections [47, 48]. For example, Herpes simplex virus (HSV-1) is a common and highly contagious virus that has been considered largely innocuous in immunocompetent individuals. Most infected people are asymptomatic but can have periodic sores in and around the mouth. However, more recent evidence has suggested that HSV-1 infection produces negative effects on human cognition [49–51] and may lead to an increase in suicidal behavior. For example, the Danish Blood Donor Study found that HSV-1 infection, as determined by seroprevalence of HSV-1 antibodies, was associated with an increased risk of suicide attempts and death [52].

*Toxoplasma gondii* (*T. gondii*), one of the most common parasites in humans, has also been associated with an increased risk of suicidal behavior. Although *T. gondii* infection has been shown to cause complications during pregnancy [53] and in immunocompromised patients [54], symptoms of infection tend to be relatively mild and time-limited in immunocompetent individuals. However, this parasite can lie dormant in brain tissue, and *T. gondii* infection is known to cause changes in host behavior [55]. *T. gondii* infection has been associated with psychiatric disorders, such as schizophrenia [56], and has been implicated in suicidal behavior [57]. In one cross-sectional, observational study, *T. gondii* seropositivity was associated with a higher rate of suicide attempts [58]. Further, when psychiatric patients with a history of suicide were compared to healthy controls, a significantly higher percentage of psychiatric patients have *T. gondii* infections [59–61]. These results have been recently confirmed by multiple meta-analyses demonstrating that *T. gondii* infection increases the risk of suicidal behavior [62, 63].

Traumatic brain injury (TBI) encompasses a wide range of injuries that can be classified as mild, moderate, or severe, depending on the extent to which consciousness and mental state are affected [64]. A variety of external forces, including blunt trauma, rapid acceleration/deceleration, penetrating injury, or blast, can cause TBI. The initial injury to brain tissue and vasculature can be localized or diffuse and is followed by a secondary injury involving changes in neurochemical and metabolic pathways, including inflammation [64]. TBI has been associated with an increased risk of psychopathology, including MDD, schizophrenia, and substance abuse [65]. TBI has also been associated with an increased risk of suicidal behavior. As early as the 1950s, it was observed that veterans returning from the first and second world wars with a TBI were committing suicide at alarming rates [66]. Since then, multiple studies have suggested an association between TBI and suicidal ideation [67], attempt [68], and death by suicide [69]. These findings were confirmed by a recent meta-analysis that found even mild TBI (i.e., concussion) was associated with a twofold risk of death by suicide [70].

Together, these results suggest that immune system activation is associated with an increase in suicidal behavior. However, this correlational observation may be influenced by a variety of factors, including the trauma associated with TBI or diagnosis with a lifelong and potentially fatal illness.

### 17.4.3 Peripheral Inflammation Observed in Suicidal Patients

Although the studies presented thus far are suggestive, a variety of confounding factors, including the presence of trauma or disease, has led researchers to examine inflammatory markers in patients experiencing suicidal behavior.

### 17.4.4 Peripheral Cytokines

One of the most common observations in suicidal patients is an increase in peripheral cytokine levels. A recent meta-analyses indicated that the pro-inflammatory cytokines IL-1β and IL-6 are the most consistently altered cytokines in the peripheral blood of suicidal patients [71]. IL-6 is produced rapidly in response to infection or injury and has a pleiotropic effect on the immune system, inducing the synthesis of acute phase proteins, promoting B- and T-cell differentiation, and inducing fever [72]. One study found that in MDD patients, those with suicidal ideation had higher levels of plasma IL-6, an effect that was independent of depression severity [73]. These findings have been confirmed by a similar study demonstrating that in patients with depression or anxiety, increased plasma IL-6 levels were associated with a higher likelihood of recent suicidal ideation, but not suicide attempts [74]. Janelidze also found increased plasma IL-6 concentrations in patients that had attempted suicide compared to both depressed patients that had not attempted suicide and healthy controls [75]. IL-6 mRNA levels in whole blood have been correlated with the severity of suicidal ideation [76]. However, others have failed to find differences in plasma IL-6 levels between suicidal and non-suicidal depressed patients [77] or in suicidal adolescents compared to age-matched healthy controls [78]. This discrepancy may relate to personality traits or method of suicide. For example, personality traits such as extraversion, impulsivity, and monotony avoidance have all been associated with an increased risk of suicidal behavior. One study found that plasma IL-6 levels were positively correlated with each of these traits in suicide attempters. Further, this study also found an association between violent methods of suicide and higher levels of plasma IL-6 [79].

The pro-inflammatory cytokine, IL-1β, has also been associated with suicide [71]. IL-1β also promotes fever and participates in activation of T cells and macrophages [80]. In patients with bipolar disorder, those at higher risk for suicide, as determined by the MINI International Neuropsychiatric Interview, had higher levels of IL-1β [81]. However, other studies have found decreased IL-1β in suicidal patients. For example, one study observed a decrease in plasma IL-1β in MDD patients that had attempted suicide within the last 5 years as compared to those that only thought about suicide and depressed patients that had not experienced suicidal ideation or made a suicide attempt [82]. The authors suggested that this may be related to the timing of sample collection but others have also found a decrease in plasma IL-1β in patients that died by suicide compared to healthy controls [83, 84]. There have also been reports that failed to find an association between suicidal behavior and IL-1β [78, 85].

In addition to IL-6 and IL-1β, other cytokines have been implicated in suicidal behavior. For example, TNF-α, another pro-inflammatory cytokine that promotes inflammation and activation of endothelial cells [80], has also been associated with suicidal behavior. TNF-α mRNA expression was significantly increased in the blood of psychiatric inpatients that had attempted suicide compared to those that had just thought about suicide [86]. This work is corroborated by another study that found increased TNF-α in the plasma of suicidal depressed patients compared to depressed patients that were not experiencing suicidal behavior [75]. TNF-α has also been positively correlated with the severity of suicidal ideation [76]. However, others have not observed a difference in TNF-α levels in the plasma of depressed patients experiencing suicidal behavior compared to those that were not suicidal [77]. Further, in adolescents, TNF-α was actually decreased in depressed patients experiencing suicidal behavior compared to depressed patients without suicidal ideation [78].

The cytokine IL-2, which promotes T-cell proliferation [80], has also been implicated in suicidal behavior though the relationship is less clear. For example, Janelidze found that plasma IL-2 levels were lower in depressed patients that had attempted suicide compared to those without suicidal behavior [75]. However, increases in the soluble IL-2 receptor have also been observed in psychiatric patients experiencing suicidal behavior compared to healthy controls [87]. Together, these results suggest that suicidal behavior is associated with changes in peripheral cytokine levels; however, the magnitude and direction of the effect may depend on a variety of factors, including the cytokine examined, the age of the patient, and specific type of suicidal behavior.

### 17.4.5 Peripheral Human C-Reactive Protein (hCRP)

Human C-reactive protein (hCRP) is an acute phase protein synthesized in the liver that is secreted in response to IL-6 or other pro-inflammatory cytokines [86, 88]. CRP plays an important role in innate immunity through its ability to activate the complement system, but its rapid and robust response to infection has also led to its use as a marker of inflammation [89]. Increases in hCRP have been associated with suicidal behavior. For example, one study found that plasma CRP levels were increased in suicide attempters, either with or without an associated syndromal depression diagnosis, compared to both controls and MDD patients not experiencing suicidal behavior [90]. This result was confirmed by Yang et al., who also found increased hCRP protein expression in the blood of depressed patients that had attempted suicide compared to both depressed patients without a previous attempt and to healthy controls [88]. Further, hCRP expression may increase with the severity of suicidal behavior as it has been shown that hCRP mRNA levels were significantly increased in patients that attempted suicide compared to those that just experience suicidal ideation [86].

Support for the association between hCRP levels and suicidal behavior also comes from larger epidemiological studies. For example, one study conducted health surveys and measured plasma hCRP levels in nearly 40,000 patients. Patients with

the highest levels of hCRP were four times more likely to die by suicide than those in the lowest group. Further, this response was graded with larger increases in serum hCRP in patients that had attempted suicide compared to those with suicidal ideation alone [91]. A cross-sectional study of over 4000 Korean adults also found that suicidal ideation was more prevalent in people with higher levels of hCRP, even after controlling for other factors including disease, depression, and BMI [92].

### 17.4.6 Kynurenine Pathway

Tryptophan is an essential amino acid that is transported into the brain and converted by the enzyme tryptophan hydroxylase to 5-hydroxytryptophan, which is then converted to 5-hydroxytryptamine (serotonin). Two major enzymes have been shown to metabolize tryptophan, tryptophan 2,3 dioxygenase (TDO), and indoleamine 2,3 dioxygenase (IDO). IDO can be directly activated by a number of pro-inflammatory cytokines, including IFNγ and TNF-α. IDO is expressed by immune cells both in the periphery and in the brain, including microglia, macrophages, and dendritic cells. IDO activation can decrease levels of tryptophan and ultimately lead to a reduction in serotonin, which has been associated with depression. Activation of IDO is also thought to have an impact on behavior through the compounds generated by the kynurenine pathway including, quinolinic acid and kynurenic acid, which act as agonists or antagonists at the NMDA receptor, respectively [23].

Multiple studies have demonstrated an association between suicidal behavior and activation of the kynurenine pathway. For example, MDD patients with a history of suicide attempt have higher plasma kynurenine concentrations than depressed patients without a history of suicide and healthy controls [93]. Suicidal patients suffering from MDD have also been shown to have lower plasma tryptophan levels and a higher kynurenine/tryptophan ratio (used as a proxy for IDO activity) than depressed patients without suicidal ideation [94]. Further, picolinic acid, a neuroprotective metabolite produced by the kynurenine pathway, is decreased in the plasma of suicide attempters [95]. Similar results have been observed in depressed adolescents displaying suicidal behavior. Suicidal adolescents had lower tryptophan levels and an elevated kynurenin/tryptophan ratio compared to depressed adolescents without suicidal behavior [96].

## 17.5 Central Nervous System Inflammation

As discussed above, multiple mechanisms exist to promote communication between the peripheral and central immune systems. Therefore, more recent studies have begun examining CNS immune activation in patients experiencing suicidal behavior.

### 17.5.1 Cerebrospinal Fluid (CSF)

Cerebrospinal fluid (CSF) bathes the brain and spinal cord and can be accessed in living patients via a lumbar puncture. Multiple studies have demonstrated cytokine changes in the CSF of patients experiencing suicidal behavior. As has been observed in the periphery, IL-6 levels are increased in the CSF of patients that attempted suicide compared to healthy controls [97, 98]. Further, CSF IL-6 levels have been positively correlated with the severity of suicidal symptoms [97]. However, another study found no difference in CSF IL-6 levels between suicide attempters and healthy controls [85]. The discrepancy in findings may be related to personality traits or the method used to attempt suicide. For example, CSF IL-6 has been associated with traits that increase suicide risk, such as monotony avoidance and impulsivity [79], as well as more violent methods of suicide [98].

IL-8 is another pro-inflammatory cytokine that has been associated with suicidal behavior. During inflammation, IL-8 acts as a chemoattractant cytokine and targets neutrophils, basophils, CD8 cell subsets, and endothelial cells [80]. One study found that IL-8 levels were decreased in the CSF of suicide attempters compared to healthy controls [85]. Another study found that female patients that attempted suicide were more likely to have a single nucleotide polymorphism (SNP) on the IL-8 gene [99]. However, others have failed to observe a difference in CSF levels of IL-8 between patients experiencing suicidal behavior and controls [97, 98]. It is possible that IL-8 levels underlie other symptoms of the psychiatric disorder rather than suicidal behavior itself. For example, when patients that had attempted suicide were divided into high or low anxiety groups, only those with high anxiety had significantly lower CSF IL-8 concentrations than controls [99].

Interestingly, some cytokines that seem to be elevated in the plasma, including TNF-α and IL-1β, are no different than healthy controls in the CSF [97, 98].

As has been observed in the periphery, multiple studies have also found alterations in the kynurenine pathway in the CSF of individuals displaying suicidal behavior. One study found that quinolinic acid, a metabolite of the kynurenine pathway that acts as an agonist of the glutamatergic NMDA receptor, was elevated in the CSF of patients that had attempted suicide compared to healthy controls. Conversely, kynurenic acid, another metabolite of the pathway that acts as an antagonist at the NMDA receptor, was not altered in suicide attempters. Interestingly, quinolinic acid levels were positively correlated with scores on the Suicide Intent Scale and were no different than controls when measured 6 months after the attempt [100]. This result was confirmed by another study that found that patients experiencing suicidal behavior had increased quinolinic acid and decreased kynurenic acid in the CSF compared to healthy controls. Further, there was a significant negative correlation between kynurenic acid levels and the severity of suicidal symptoms [97].

### 17.5.2 Brain Imaging

Although measuring inflammation in the living brain remains a challenge, the 18-kDa translocator protein (TSPO) has been used as a proxy for measuring neuroinflammation. TSPO is a mitochondrial protein that can be measured in the brain using positron emission tomography (PET) imaging. In the healthy brain, TSPO binding remains relatively low but is upregulated during neuropathological conditions [101]. Although TSPO upregulation was originally thought to occur primarily in microglia, recent work suggests that it may also represent local myeloid cell proliferation and monocyte infiltration in to the brain [102]. Increased TSPO binding has been observed in the brains of patients suffering from major depressive disorder [103–105]. TSPO binding has also been measured in suicidal patients, although few studies have been performed and the findings remain inconclusive. One study found that MDD patients with suicidal thoughts had greater TSPO binding in the anterior cingulate cortex (ACC) compared to healthy controls or depressed patients without suicidal ideation [104]. However, a subsequent study found that although depressed patients had elevated TSPO binding in the ACC compared to controls, there were no differences in depressed patients with and without suicidal behavior [105].

### 17.5.3 Postmortem Brain

The only way to directly measure inflammation in the brain is in postmortem tissue, and multiple studies have examined cytokines in the postmortem brain of suicide victims. Some studies have demonstrated that cytokines that are elevated in plasma and CSF of suicidal patients are also increased in the brain. IL-6, IL-1β, and TNF-α, for example, are all increased in the prefrontal cortex (Broadman's Area 10) of teenagers that died by suicide compared to people that died by other means [106]. Others have observed increased IL-6, IL-1β, and TNF-α mRNA expression in another prefrontal region (Broadman's Area 9) in depressed patients that died by suicide compared to controls without a psychiatric diagnosis [107]. The increase in IL-6 expression was also observed when tissue was pooled from multiple regions, including the amygdala, gyrus cinguli, hippocampus, and pons [108]. However, other studies have failed to identify a difference in IL-6, IL-1β, and TNF-α mRNA expression in the ventrolateral prefrontal cortex between depressed patients that died by suicide compared to depressed patients that died by other means [109]. Further, no difference in IL-6, IL-1β, or TNF-α mRNA expression was observed in the orbital frontal cortex (Broadman's Area 11) of suicide victims [110], suggesting that cytokine alterations may be restricted to specific brain regions.

Examination of additional cytokines has also identified differences between suicide victims and controls. For example, Tonelli et al. found that female suicide victims had higher levels of IL-4 in the orbitofrontal cortex compared to controls [110]. IL-4 is a pro-inflammatory cytokine that induces activation of B cells and promotes differentiation toward Th2 cells [80]. At the same time, male suicide

victims had higher levels of IL-13 in the orbitofrontal cortex than controls [110]. This cytokine induces B-cell growth and differentiation, inhibits cytokine production by macrophages and Th1 cells, and induces allergy and asthma [80]. Further, in pooled tissue from the amygdala, gyrus cinguli, hippocampus, and pons, suicide victims had increased expression of interferon-associated genes, including IFNA1, IFNA2, IFNB1, and IFNG, genes associated with Toll-like receptors (TLRs), including TLR3, TLR7, and TLR8, and cytokine genes, such as TIMP-1 and CXCL9 [108]. Others have also observed a change in TLRs, which can be activated my PAMPs and DAMPs, ultimately leading to the production of cytokines through the activation of the transcription factor, NFkB. Pandey et al. found that depressed patients that died by suicide had an increase in TLR2, TLR3, TLR4, TLR6, and TLR10 protein expression in the prefrontal cortex compared to control subjects [111]. These results suggest that additional cytokines may play a role in the central inflammation associated with suicide.

In addition to altered cytokine expression, changes in the resident immune cells, microglia, have also been observed in the brains of suicide victims. The idea that microglia may be altered in suicide victims was first proposed in 2006 when human leukocyte antigen–DR isotype (HLA-DR) staining was used to label microglia in postmortem brains of schizophrenia patients. Two schizophrenic patients had a marked increase in microglia cell number in the anterior cingulate cortex (ACC) and mediodorsal thalamus (MDT) compared to the other samples. These two patients had died by suicide [112]. Subsequently, Steiner et al. compared HLA-DR staining in the dorsolateral prefrontal cortex (DLPFC), ACC, MD, and hippocampus of healthy controls, patients with schizophrenia and depression. Although diagnosis per se (depression or schizophrenia) had no effect on microglia density, microgliosis was observed in the DLPFC, ACC, and MD of patients that died by suicide. Another study that used ionized calcium-binding adaptor molecule 1 (Iba1) to label microglia did not find a difference between the total number of microglia or the ratio of primed versus ramified microglia in suicide victims compared to controls. However, this study did find that suicide victims had more blood vessels with a high density of Iba1-positive cells surrounding them than controls. Further, gene expression of IBA1, CD45, and MCP-1 were significantly increased in suicide victims compared to controls [113]. In addition, microglia have been examined in other brain regions, including the dorsal raphe nucleus, the predominant serotonergic cell group in the brainstem. HLD-DR staining was used to show that depressed patients that died by suicide had significantly more microglial reactivity than depressed patients without suicidal thoughts [114]. Alterations in the kynurenine pathway within microglia have also been observed in suicide victims; however, the result seems to be contrary to that observed in the periphery and CSF. Depressed patients that died by suicide had fewer hippocampal microglia expressing quinolinic acid compared to controls [115].

Astrocytes also play a role in neuroimmune signaling, and there is some evidence to suggest that alterations in astrocyte function are associated with suicidal behavior. In patients that died by suicide, astrocytes located in white matter adjacent to the anterior cingulate cortex (Broadman's Area 24) had larger cell bodies and more

ramified processes than those found in control brains [116]. A decrease in the number of astrocytes labeled with glial fibrillary acidic protein (GFAP) has also been observed in the prefrontal cortex, caudate nucleus, and mediodorsal thalamus of depressed patients that died by suicide compared to controls [117]. This effect was also observed when other astrocytic markers, including vimentin, which labels different populations of astrocytes were used [117]. Further, in depressed patients that died by suicide, the area of astrocyte staining with GFAP was inversely correlated with the duration of depression [118].

## 17.6 Treatment Studies

While very few studies have examined the effect of anti-inflammatory treatments on suicidal behavior directly, there is increasing evidence that reducing inflammation may be a useful treatment strategy for disorders associated with suicide, including depression. For example, drugs that are currently being used as antidepressants, and are known to reduce suicidal behavior, have been shown to possess anti-inflammatory properties. Multiple meta-analyses have demonstrated a reduction in peripheral cytokines, including IL-6, TNF-α, IL-10, and CCL2, after treatment with antidepressants [119–124]. However, in at least one analysis, the reduction in cytokines was not correlated with treatment response [119]. The results of these studies are fairly heterogeneous, with multiple factors contributing to variable results, including baseline inflammation, BMI, smoking status, and type of antidepressant used [125]. For example, selective serotonin reuptake inhibitors (SSRIs) and cognitive behavioral therapy have both been shown to reduce peripheral inflammation [123, 126]. Conversely, serotonin and norepinephrine reuptake inhibitors (SNRIs) and ECT have been shown to induce cytokines [120, 127, 128]. One recent study examined peripheral inflammation in response to ketamine, a glutamatergic NMDA receptor antagonist that has been shown to rapidly reduce symptoms of depression, including suicidal ideation [129, 130]. Circulating monocytes, the main source of macrophages that infiltrate tissue during infection, were collected from MDD patients that had been hospitalized for suicidal ideation or attempt. The study demonstrated that in vitro treatment with ketamine biased monocytes toward an M2c phenotype, with lower expression of HLA-DR and other activation markers. Further, ketamine-treated monocytes had lower baseline IL-6 and IL-10 production and produced less TNF-α when challenged with lipopolysaccharide [131].

In addition to studying the effect of established antidepressants on immune factors, others have examined the utility of anti-inflammatory treatments in depression. Nonsteroidal anti-inflammatory drugs (NSAIDs), for example, have been shown to be useful both as an add-on therapy to traditional antidepressants and as a stand-alone treatment strategy [132]. NSAID treatment has also been associated with a reduction in suicidal ideation. Using data provided by MedWatch and the Food and Drug Administration Safety Information and Adverse Event Reporting Program, significantly less suicidal ideation was reported in patients treated with NSAIDs compared to acetaminophen (a non-NSAID) [133]. In addition, cytokine

inhibitor therapies have also been examined as a potential treatment strategy for depression [132, 134], although it should be noted that most studies have been conducted in patients with comorbid inflammatory disorders [46, 135–141]. In depressed patients without a comorbid inflammatory disorder, infliximab, a TNF-α-neutralizing antibody, seems to be effective, but only in a subset of treatment-resistant patients with elevated baseline levels of inflammation [142]. This drug has also been shown to reduce depression symptoms in patients that experienced childhood trauma [143], an effect that was associated with its anti-inflammatory properties [144].

## 17.7 Potential Mechanism: Early-Life stress

There is strong evidence that stressful experiences in early life increase risk of developing a spectrum of diseases in adulthood, including major psychiatric and other medical disorders. This is particularly concerning considering that in the United States in 2012, 3.4 million cases of child abuse and neglect were referred to child protective services, representing 686,000 children [145]. The vast majority of cases never get reported to law enforcement and self-report studies suggest that up to 40% of the population experience some form of neglect or abuse [146, 147]. Although clinical observations have long suggested a connection between early-life stress and adult psychopathology [148], the 1998 Adverse Childhood Experiences (ACE) study sponsored by the US Center for Disease Control, provided more conclusive evidence that early life stress can lead to psychiatric disorders. In the ACE study, a standardized medical exam and questionnaire about early-life experiences were administered to >10,000 individuals living in the San Diego area. The study compared the number of ACEs experienced, including psychological, physical or sexual abuse, domestic violence, household substance abuse, and parental loss, with overall health. They found that over 50% of respondents had experienced at least one category of childhood exposure, and about 25% experienced two or more ACEs. Further, the study demonstrated a significant association between ACEs and disease. People that experience four or more ACEs had an increased risk for developing psychiatric disorders, including depression, anxiety, panic attacks, substance, and alcohol abuse. Alarmingly, the ACE Study found that people who had experienced four or more ACEs also had a 12-fold risk of suicidal behavior [149]. This risk of suicidal behavior remains even when other risk factors are controlled, including alcoholism, drug use, or depression [150]. Further, risk of suicidal behavior increases as the number of ACEs increases, with each additional ACE increasing the risk of suicide attempt by about 60% [150].

The relationship between suicide and early-life stress has been repeatedly confirmed in numerous studies, examining a variety of early life stressors [151, 152]. For example, childhood sexual abuse has consistently been shown to increase suicide risk [153–156], which was demonstrated by a recent meta-analysis that found that childhood sexual abuse causes a fourfold increase in the risk of attempting suicide [157]. Physical abuse and bullying have also been associated with

an increased risk of suicidal behavior [158]. Certain factors, including the frequency of the abuse, sex of the victim, and identity of the abuser, may influence the likelihood of suicidal behavior [159, 160].

Interestingly, the ACE Study found that early-life stress was associated with not only psychiatric diseases and suicidal behavior but also other negative health outcomes, including sleep disturbances, obesity, smoking, COPD, asthma, and heart disease [149]. Similar to the relationship between early-life stress and psychiatric disorders and suicide, the risk of developing another medical disorder is proportional to the number and magnitude of ACEs [149, 161–164]. Interestingly, inflammation has been associated with or is implicated in the pathology of many of these medical diagnoses, suggesting that the immune system may be one mechanism by which early-life stress may lead to psychiatric and other medical disorders. Over the past 30 years, several studies have demonstrated that stress can alter immune function. Acute stressors lead to activation of the hypothalamic-pituitary-adrenal (HPA) axis and the sympathetic nervous system (SNS), which promote physiological responses that help deal with the threat. These physiological changes are mediated in part by the release of hormones, including glucocorticoids and corticotropin-releasing hormone. These stress hormones can influence the immune system by binding to receptors on immune cells or indirectly [165, 166]. While this response may be adaptive in adults, evidence suggests that stress during development may lead to long-term changes in immune function [167]. For example, one study found that a standard laboratory stress test produced an elevated IL-6 response in depressed men with a history of childhood abuse or neglect compared to controls [168]. One of the first human studies to examine the relationship between early-life stress and immune function examined hCRP levels and white blood cell numbers in adults that had experienced early-life stress. They found that adults that had experience childhood maltreatment were 1.5 times more likely to have elevated hCRP levels and an increased number of white blood cells. This result was independent of cooccuring childhood risk factors, including low birth weight and socioeconomic disadvantages, adult stress, and participation in health-damaging behaviors [169]. In adult patients with depression, the elevation in hCRP seems to be driven by early-life stress as depressed patients without early life trauma did not have elevated hCRP levels [170]. In breast cancer survivors, early life stress was linked to IL-6 levels, even when controlling for treatment, age, BMI, ethnicity, and alcohol use [171]. Further, the timing of the stressor may play a role as stressors experienced in middle childhood (6–8 years) were associated with higher levels of peripheral IL-6 and CRP at age 10 and 15, although these effects were at least partially mediated by BMI [172]. Together, these results suggest that one biological mechanism by which early-life stress may influence the risk of suicidal behavior is via the immune system. Future work should determine the utility of immune factors as biomarkers for suicide risk and determine whether targeting the immune system can reduce suicidal behavior, especially in at-risk groups, such as those that have experienced early-life stress.

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## References


1. World Health Organization. Suicide worldwide in 2019: global health estimates. Geneva: World Health Organization; 2019.
2. Hedegaard H, Curtin SC, Warner M. Suicide mortality in the United States, 1999-2019; 2021.
3. Shepard DS, Gurewich D, Lwin AK, Reed GA Jr, Silverman MM. Suicide and suicidal attempts in the United States: costs and policy implications. Suicide Life Threat Behav. 2016;46(3):352–62.
4. National strategy for suicide prevention: goals and objectives for action: a report of the U.S. Surgeon General and of the National Action Alliance for Suicide Prevention, 2012. Center for Disease Control; 2012.
5. World Health Organization. Preventing suicide: a global imperative. Geneva: World Health Organization; 2014.
6. Nock MK, Deming CA, Fullerton CS, Gilman SE, Goldenberg M, Kessler RC, et al. Suicide among soldiers: a review of psychosocial risk and protective factors. Psychiatry. 2013;76(2): 97–125.
7. Carroll R, Metcalfe C, Gunnell D. Hospital presenting self-harm and risk of fatal and non-fatal repetition: systematic review and meta-analysis. PLoS One. 2014;9(2):e89944.
8. Arsenault-Lapierre G, Kim C, Turecki G. Psychiatric diagnoses in 3275 suicides: a meta-analysis. BMC Psychiatry. 2004;4:37.
9. McGirr A, Renaud J, Seguin M, Alda M, Turecki G. Course of major depressive disorder and suicide outcome: a psychological autopsy study. J Clin Psychiatry. 2008;69(6):966–70.
10. Holma KM, Melartin TK, Haukka J, Holma IA, Sokero TP, Isometsa ET. Incidence and predictors of suicide attempts in DSM-IV major depressive disorder: a five-year prospective study. Am J Psychiatry. 2010;167(7):801–8.
11. Hor K, Taylor M. Suicide and schizophrenia: a systematic review of rates and risk factors. J Psychopharmacol. 2010;24(4 Suppl):81–90.
12. McGirr A, Alda M, Seguin M, Cabot S, Lesage A, Turecki G. Familial aggregation of suicide explained by cluster B traits: a three-group family study of suicide controlling for major depressive disorder. Am J Psychiatry. 2009;166(10):1124–34.
13. Murphy K, Weaver C. Janeway's immunobiology. New York: Garland Science; 2012.
14. MacGillivray DM, Kollmann TR. The role of environmental factors in modulating immune responses in early life. Front Immunol. 2014;5:434.
15. Merle NS, Church SE, Fremeaux-Bacchi V, Roumenina LT. Complement system part I - molecular mechanisms of activation and regulation. Front Immunol. 2015;6:262.

16. Galea I, Bechmann I, Perry VH. What is immune privilege (not)? Trends Immunol. 2007;28 (1):12–8.
17. Nimmerjahn A, Kirchhoff F, Helmchen F. Resting microglial cells are highly dynamic surveillants of brain parenchyma in vivo. Science. 2005;308(5726):1314–8.
18. Salter MW, Stevens B. Microglia emerge as central players in brain disease. Nat Med. 2017;23 (9):1018–27.
19. Saijo K, Glass CK. Microglial cell origin and phenotypes in health and disease. Nat Rev Immunol. 2011;11(11):775–87.
20. Tan CX, Eroglu C. Cell adhesion molecules regulating astrocyte-neuron interactions. Curr Opin Neurobiol. 2021;69:170–7.
21. Liddelow SA, Guttenplan KA, Clarke LE, Bennett FC, Bohlen CJ, Schirmer L, et al. Neurotoxic reactive astrocytes are induced by activated microglia. Nature. 2017;541(7638):481–7.
22. Ballabh P, Braun A, Nedergaard M. The blood-brain barrier: an overview: structure, regulation, and clinical implications. Neurobiol Dis. 2004;16(1):1–13.
23. Dantzer R, O'Connor JC, Freund GG, Johnson RW, Kelley KW. From inflammation to sickness and depression: when the immune system subjugates the brain. Nat Rev Neurosci. 2008;9(1):46–56.
24. Ganong WF. Circumventricular organs: definition and role in the regulation of endocrine and autonomic function. Clin Exp Pharmacol Physiol. 2000;27(5–6):422–7.
25. Vitkovic L, Konsman JP, Bockaert J, Dantzer R, Homburger V, Jacque C. Cytokine signals propagate through the brain. Mol Psychiatry. 2000;5(6):604–15.
26. Konsman JP, Vigues S, Mackerlova L, Bristow A, Blomqvist A. Rat brain vascular distribution of interleukin-1 type-1 receptor immunoreactivity: relationship to patterns of inducible cyclooxygenase expression by peripheral inflammatory stimuli. J Comp Neurol. 2004;472(1): 113–29.
27. Schiltz JC, Sawchenko PE. Distinct brain vascular cell types manifest inducible cyclooxygenase expression as a function of the strength and nature of immune insults. J Neurosci. 2002;22 (13):5606–18.
28. Berraondo P, Sanmamed MF, Ochoa MC, Etxeberria I, Aznar MA, Perez-Gracia JL, et al. Cytokines in clinical cancer immunotherapy. Br J Cancer. 2019;120(1):6–15.
29. Fyfe G, Fisher RI, Rosenberg SA, Sznol M, Parkinson DR, Louie AC. Results of treatment of 255 patients with metastatic renal cell carcinoma who received high-dose recombinant interleukin-2 therapy. J Clin Oncol. 1995;13(3):688–96.
30. Atkins MB, Lotze MT, Dutcher JP, Fisher RI, Weiss G, Margolin K, et al. High-dose recombinant interleukin 2 therapy for patients with metastatic melanoma: analysis of 270 patients treated between 1985 and 1993. J Clin Oncol. 1999;17(7):2105–16.
31. Musselman DL, Lawson DH, Gumnick JF, Manatunga AK, Penna S, Goodkin RS, et al. Paroxetine for the prevention of depression induced by high-dose interferon alfa. N Engl J Med. 2001;344(13):961–6.
32. Renault PF, Hoofnagle JH, Park Y, Mullen KD, Peters M, Jones DB, et al. Psychiatric complications of long-term interferon alfa therapy. Arch Intern Med. 1987;147(9):1577–80.
33. Fragoso YD, Frota ER, Lopes JS, Noal JS, Giacomo MC, Gomes S, et al. Severe depression, suicide attempts, and ideation during the use of interferon beta by patients with multiple sclerosis. Clin Neuropharmacol. 2010;33(6):312–6.
34. Janssen HL, Brouwer JT, van der Mast RC, Schalm SW. Suicide associated with alfa-interferon therapy for chronic viral hepatitis. J Hepatol. 1994;21(2):241–3.
35. Baron DA, Hardie T, Baron SH. Possible association of interleukin-2 treatment with depression and suicide. J Am Osteopath Assoc. 1993;93(7):799–800.
36. Rifflet H, Vuillemin E, Oberti F, Duverger P, Lainé P, Garré JB, et al. [Suicidal impulses in patients with chronic viral hepatitis C during or after therapy with interferon alpha]. Gastroenterol Clin Biol. 1998;22(3):353–7.

37. Capuron L, Ravaud A, Miller AH, Dantzer R. Baseline mood and psychosocial characteristics of patients developing depressive symptoms during interleukin-2 and/or interferon-alpha cancer therapy. Brain Behav Immun. 2004;18(3):205–13.
38. Dieperink E, Ho SB, Tetrick L, Thuras P, Dua K, Willenbring ML. Suicidal ideation during interferon-alpha2b and ribavirin treatment of patients with chronic hepatitis C. Gen Hosp Psychiatry. 2004;26(3):237–40.
39. Stenager EN, Stenager E, Koch-Henriksen N, Brønnum-Hansen H, Hyllested K, Jensen K, et al. Suicide and multiple sclerosis: an epidemiological investigation. J Neurol Neurosurg Psychiatry. 1992;55(7):542–5.
40. Sadovnick AD, Eisen K, Ebers GC, Paty DW. Cause of death in patients attending multiple sclerosis clinics. Neurology. 1991;41(8):1193–6.
41. Pompili M, Innamorati M, Trovarelli S, Narcisi A, Bellini S, Orsini D, et al. Suicide risk and psychiatric comorbidity in patients with psoriasis. J Int Med Res. 2016;44(1 Suppl):61–6.
42. Gupta MA, Schork NJ, Gupta AK, Kirkby S, Ellis CN. Suicidal ideation in psoriasis. Int J Dermatol. 1993;32(3):188–90.
43. Picardi A, Lega I, Tarolla E. Suicide risk in skin disorders. Clin Dermatol. 2013;31(1):47–56.
44. Kurd SK, Troxel AB, Crits-Christoph P, Gelfand JM. The risk of depression, anxiety, and suicidality in patients with psoriasis: a population-based cohort study. Arch Dermatol. 2010;146(8):891–5.
45. Lebwohl MG, Papp KA, Marangell LB, Koo J, Blauvelt A, Gooderham M, et al. Psychiatric adverse events during treatment with brodalumab: analysis of psoriasis clinical trials. J Am Acad Dermatol. 2018;78(1):81–9.e5.
46. Griffiths CEM, Fava M, Miller AH, Russell J, Ball SG, Xu W, et al. Impact of ixekizumab treatment on depressive symptoms and systemic inflammation in patients with moderate-to-severe psoriasis: an integrated analysis of three phase 3 clinical studies. Psychother Psychosom. 2017;86(5):260–7.
47. Lund-Sørensen H, Benros ME, Madsen T, Sørensen HJ, Eaton WW, Postolache TT, et al. A nationwide cohort study of the association between hospitalization with infection and risk of death by suicide. JAMA Psychiatry. 2016;73(9):912–9.
48. Dickerson F, Wilcox HC, Adamos M, Katsafanas E, Khushalani S, Origoni A, et al. Suicide attempts and markers of immune response in individuals with serious mental illness. J Psychiatr Res. 2017;87:37–43.
49. Dickerson F, Stallings C, Origoni A, Vaughan C, Khushalani S, Yolken R. Additive effects of elevated C-reactive protein and exposure to herpes simplex virus type 1 on cognitive impairment in individuals with schizophrenia. Schizophr Res. 2012;134(1):83–8.
50. Schretlen DJ, Vannorsdall TD, Winicki JM, Mushtaq Y, Hikida T, Sawa A, et al. Neuroanatomic and cognitive abnormalities related to herpes simplex virus type 1 in schizophrenia. Schizophr Res. 2010;118(1–3):224–31.
51. Yolken RH, Torrey EF, Lieberman JA, Yang S, Dickerson FB. Serological evidence of exposure to herpes simplex virus type 1 is associated with cognitive deficits in the CATIE schizophrenia sample. Schizophr Res. 2011;128(1-3):61–5.
52. Nissen J, Trabjerg B, Pedersen MG, Banasik K, Pedersen OB, Sørensen E, et al. Herpes simplex virus type 1 infection is associated with suicidal behavior and first registered psychiatric diagnosis in a healthy population. Psychoneuroendocrinology. 2019;108:150–4.
53. Carellos EV, de Andrade GM, Vasconcelos-Santos DV, Januário JN, Romanelli RM, Abreu MN, et al. Adverse socioeconomic conditions and oocyst-related factors are associated with congenital toxoplasmosis in a population-based study in Minas Gerais, Brazil. PLoS One. 2014;9(2):e88588.
54. Ayi I, Sowah AO, Blay EA, Suzuki T, Ohta N, Ayeh-Kumi PF. Toxoplasma gondii infections among pregnant women, children and HIV-seropositive persons in Accra, Ghana. Trop Med Health. 2016;44:17.
55. Tong WH, Pavey C, O'Handley R, Vyas A. Behavioral biology of Toxoplasma gondii infection. Parasit Vectors. 2021;14(1):77.

56. Chaudhury A, Ramana BV. Schizophrenia and bipolar disorders: the toxoplasma connection. Trop Parasitol. 2019;9(2):71–6.
57. Postolache TT, Wadhawan A, Rujescu D, Hoisington AJ, Dagdag A, Baca-Garcia E, et al. Toxoplasma gondii, suicidal behavior, and intermediate phenotypes for suicidal behavior. Front Psychiatry. 2021;12:665682.
58. Zhang Y, Träskman-Bendz L, Janelidze S, Langenberg P, Saleh A, Constantine N, et al. Toxoplasma gondii immunoglobulin G antibodies and nonfatal suicidal self-directed violence. J Clin Psychiatry. 2012;73(8):1069–76.
59. Bak J, Shim SH, Kwon YJ, Lee HY, Kim JS, Yoon H, et al. The association between suicide attempts and Toxoplasma gondii infection. Clin Psychopharmacol Neurosci. 2018;16(1): 95–102.
60. Yagmur F, Yazar S, Temel HO, Cavusoglu M. May Toxoplasma gondii increase suicide attempt-preliminary results in Turkish subjects? Forensic Sci Int. 2010;199(1–3):15–7.
61. Ling VJ, Lester D, Mortensen PB, Langenberg PW, Postolache TT. Toxoplasma gondii seropositivity and suicide rates in women. J Nerv Ment Dis. 2011;199(7):440–4.
62. Soleymani E, Faizi F, Heidarimoghadam R, Davoodi L, Mohammadi Y. Association of T. gondii infection with suicide: a systematic review and meta-analysis. BMC Public Health. 2020;20(1):766.
63. Amouei A, Moosazadeh M, Nayeri Chegeni T, Sarvi S, Mizani A, Pourasghar M, et al. Evolutionary puzzle of Toxoplasma gondii with suicidal ideation and suicide attempts: an updated systematic review and meta-analysis. Transbound Emerg Dis. 2020.
64. Meier TB, Savitz J. The Kynurenine pathway in traumatic brain injury: implications for psychiatric outcomes. Biol Psychiatry. 2022;91(5):449–58.
65. Holzer KJ, Carbone JT, DeLisi M, Vaughn MG. Traumatic brain injury and coextensive psychopathology: new evidence from the 2016 Nationwide Emergency Department Sample (NEDS). J Psychiatr Res. 2019;114:149–52.
66. Russell WR. Disability caused by brain wounds; a review of 1,166 cases. J Neurol Neurosurg Psychiatry. 1951;14(1):35–9.
67. Anstey KJ, Butterworth P, Jorm AF, Christensen H, Rodgers B, Windsor TD. A population survey found an association between self-reports of traumatic brain injury and increased psychiatric symptoms. J Clin Epidemiol. 2004;57(11):1202–9.
68. Silver JM, Kramer R, Greenwald S, Weissman M. The association between head injuries and psychiatric disorders: findings from the New Haven NIMH Epidemiologic Catchment Area Study. Brain Inj. 2001;15(11):935–45.
69. Teasdale TW, Engberg AW. Suicide after traumatic brain injury: a population study. J Neurol Neurosurg Psychiatry. 2001;71(4):436–40.
70. Fralick M, Sy E, Hassan A, Burke MJ, Mostofsky E, Karsies T. Association of concussion with the risk of suicide: a systematic review and meta-analysis. JAMA Neurol. 2019;76(2): 144–51.
71. Black C, Miller BJ. Meta-analysis of cytokines and chemokines in suicidality: distinguishing suicidal versus nonsuicidal patients. Biol Psychiatry. 2015;78(1):28–37.
72. Tanaka T, Narazaki M, Kishimoto T. IL-6 in inflammation, immunity, and disease. Cold Spring Harb Perspect Biol. 2014;6(10):a016295.
73. O'Donovan A, Rush G, Hoatam G, Hughes BM, McCrohan A, Kelleher C, et al. Suicidal ideation is associated with elevated inflammation in patients with major depressive disorder. Depress Anxiety. 2013;30(4):307–14.
74. Dolsen MR, Prather AA, Lamers F, Penninx B. Suicidal ideation and suicide attempts: associations with sleep duration, insomnia, and inflammation. Psychol Med. 2020:1–10.
75. Janelidze S, Mattei D, Westrin Å, Träskman-Bendz L, Brundin L. Cytokine levels in the blood may distinguish suicide attempters from depressed patients. Brain Behav Immun. 2011;25(2): 335–9.

76. Rengasamy M, Zhong Y, Marsland A, Chen K, Douaihy A, Brent D, et al. Signaling networks in inflammatory pathways and risk for suicidal behavior. Brain Behav Immun Health. 2020;7:100122.
77. Karlović D, Serretti A, Vrkić N, Martinac M, Marčinko D. Serum concentrations of CRP, IL-6, TNF-α and cortisol in major depressive disorder with melancholic or atypical features. Psychiatry Res. 2012;198(1):74–80.
78. Gabbay V, Klein RG, Guttman LE, Babb JS, Alonso CM, Nishawala M, et al. A preliminary study of cytokines in suicidal and nonsuicidal adolescents with major depression. J Child Adolesc Psychopharmacol. 2009;19(4):423–30.
79. Isung J, Aeinehband S, Mobarrez F, Nordström P, Runeson B, Asberg M, et al. High interleukin-6 and impulsivity: determining the role of endophenotypes in attempted suicide. Transl Psychiatry. 2014;4(10):e470.
80. Köhler CA, Freitas TH, Maes M, de Andrade NQ, Liu CS, Fernandes BS, et al. Peripheral cytokine and chemokine alterations in depression: a meta-analysis of 82 studies. Acta Psychiatr Scand. 2017;135(5):373–87.
81. Monfrim X, Gazal M, De Leon PB, Quevedo L, Souza LD, Jansen K, et al. Immune dysfunction in bipolar disorder and suicide risk: is there an association between peripheral corticotropin-releasing hormone and interleukin-1β? Bipolar Disord. 2014;16(7):741–7.
82. Ganança L, Galfalvy HC, Cisneros-Trujillo S, Basseda Z, Cooper TB, Ren X, et al. Relationships between inflammatory markers and suicide risk status in major depression. J Psychiatr Res. 2021;134:192–9.
83. Lu J, Li S, Li H, Mou T, Zhou L, Huang B, et al. Changes in plasma NPY, IL-1β and hypocretin in people who died by suicide. Neuropsychiatr Dis Treat. 2019;15:2893–900.
84. Coryell W, Wilcox H, Evans SJ, Pandey GN, Jones-Brando L, Dickerson F, et al. Aggression, impulsivity and inflammatory markers as risk factors for suicidal behavior. J Psychiatr Res. 2018;106:38–42.
85. Isung J, Mobarrez F, Nordström P, Asberg M, Jokinen J. Low plasma vascular endothelial growth factor (VEGF) associated with completed suicide. World J Biol Psychiatry. 2012;13(6):468–73.
86. Melhem NM, Munroe S, Marsland A, Gray K, Brent D, Porta G, et al. Blunted HPA axis activity prior to suicide attempt and increased inflammation in attempters. Psychoneuroendocrinology. 2017;77:284–94.
87. Nässberger L, Träskman-Bendz L. Increased soluble interleukin-2 receptor concentrations in suicide attempters. Acta Psychiatr Scand. 1993;88(1):48–52.
88. Yang Y, Chen J, Liu C, Fang L, Liu Z, Guo J, et al. The extrinsic coagulation pathway: a biomarker for suicidal behavior in major depressive disorder. Sci Rep. 2016;6:32882.
89. Dupuy AM, Terrier N, Sénécal L, Morena M, Leray H, Canaud B, et al. [Is C-reactive protein a marker of inflammation?]. Nephrologie. 2003;24(7):337–41.
90. Ventorp F, Gustafsson A, Träskman-Bendz L, Westrin Å, Ljunggren L. Increased soluble urokinase-type plasminogen activator receptor (suPAR) levels in plasma of suicide attempters. PLoS One. 2015;10(10):e0140052.
91. Gibbs HM, Davis L, Han X, Clothier J, Eads LA, Cáceda R. Association between C-reactive protein and suicidal behavior in an adult inpatient population. J Psychiatr Res. 2016;79:28–33.
92. Park RJ, Kim YH. Association between high sensitivity CRP and suicidal ideation in the Korean general population. Eur Neuropsychopharmacol. 2017;27(9):885–91.
93. Sublette ME, Galfalvy HC, Fuchs D, Lapidus M, Grunebaum MF, Oquendo MA, et al. Plasma kynurenine levels are elevated in suicide attempters with major depressive disorder. Brain Behav Immun. 2011;25(6):1272–8.
94. Messaoud A, Mensi R, Douki W, Neffati F, Najjar MF, Gobbi G, et al. Reduced peripheral availability of tryptophan and increased activation of the kynurenine pathway and cortisol correlate with major depression and suicide. World J Biol Psychiatry. 2019;20(9):703–11.

95. Brundin L, Sellgren CM, Lim CK, Grit J, Pålsson E, Landén M, et al. An enzyme in the kynurenine pathway that governs vulnerability to suicidal behavior by regulating excitotoxicity and neuroinflammation. Transl Psychiatry. 2016;6(8):e865.
96. Bradley KA, Case JA, Khan O, Ricart T, Hanna A, Alonso CM, et al. The role of the kynurenine pathway in suicidality in adolescent major depressive disorder. Psychiatry Res. 2015;227(2–3):206–12.
97. Bay-Richter C, Linderholm KR, Lim CK, Samuelsson M, Träskman-Bendz L, Guillemin GJ, et al. A role for inflammatory metabolites as modulators of the glutamate N-methyl-D-aspartate receptor in depression and suicidality. Brain Behav Immun. 2015;43:110–7.
98. Lindqvist D, Janelidze S, Hagell P, Erhardt S, Samuelsson M, Minthon L, et al. Interleukin-6 is elevated in the cerebrospinal fluid of suicide attempters and related to symptom severity. Biol Psychiatry. 2009;66(3):287–92.
99. Janelidze S, Suchankova P, Ekman A, Erhardt S, Sellgren C, Samuelsson M, et al. Low IL-8 is associated with anxiety in suicidal patients: genetic variation and decreased protein levels. Acta Psychiatr Scand. 2015;131(4):269–78.
100. Erhardt S, Lim CK, Linderholm KR, Janelidze S, Lindqvist D, Samuelsson M, et al. Connecting inflammation with glutamate agonism in suicidality. Neuropsychopharmacology. 2013;38(5):743–52.
101. Rupprecht R, Papadopoulos V, Rammes G, Baghai TC, Fan J, Akula N, et al. Translocator protein (18 kDa) (TSPO) as a therapeutic target for neurological and psychiatric disorders. Nat Rev Drug Discov. 2010;9(12):971–88.
102. Owen DR, Narayan N, Wells L, Healy L, Smyth E, Rabiner EA, et al. Pro-inflammatory activation of primary microglia and macrophages increases 18 kDa translocator protein expression in rodents but not humans. J Cereb Blood Flow Metab. 2017;37(8):2679–90.
103. Setiawan E, Wilson AA, Mizrahi R, Rusjan PM, Miler L, Rajkowska G, et al. Role of translocator protein density, a marker of neuroinflammation, in the brain during major depressive episodes. JAMA Psychiatry. 2015;72(3):268–75.
104. Holmes SE, Hinz R, Conen S, Gregory CJ, Matthews JC, Anton-Rodriguez JM, et al. Elevated translocator protein in anterior cingulate in major depression and a role for inflammation in suicidal thinking: a positron emission tomography study. Biol Psychiatry. 2018;83(1):61–9.
105. Schubert JJ, Veronese M, Fryer TD, Manavaki R, Kitzbichler MG, Nettis MA, et al. A modest increase in (11)C-PK11195-positron emission tomography TSPO binding in depression is not associated with serum C-reactive protein or body mass index. Biol Psychiatry Cogn Neurosci Neuroimaging. 2021;6(7):716–24.
106. Pandey GN, Rizavi HS, Ren X, Fareed J, Hoppensteadt DA, Roberts RC, et al. Proinflammatory cytokines in the prefrontal cortex of teenage suicide victims. J Psychiatr Res. 2012;46(1):57–63.
107. Pandey GN, Rizavi HS, Zhang H, Bhaumik R, Ren X. Abnormal protein and mRNA expression of inflammatory cytokines in the prefrontal cortex of depressed individuals who died by suicide. J Psychiatry Neurosci. 2018;43(6):376–85.
108. Hoyo-Becerra C, Huebener A, Trippler M, Lutterbeck M, Liu ZJ, Truebner K, et al. Concomitant interferon alpha stimulation and TLR3 activation induces neuronal expression of depression-related genes that are elevated in the brain of suicidal persons. PLoS One. 2013;8(12):e83149.
109. Clark SM, Pocivavsek A, Nicholson JD, Notarangelo FM, Langenberg P, McMahon RP, et al. Reduced kynurenine pathway metabolism and cytokine expression in the prefrontal cortex of depressed individuals. J Psychiatry Neurosci. 2016;41(6):386–94.
110. Tonelli LH, Stiller J, Rujescu D, Giegling I, Schneider B, Maurer K, et al. Elevated cytokine expression in the orbitofrontal cortex of victims of suicide. Acta Psychiatr Scand. 2008;117(3):198–206.
111. Pandey GN, Rizavi HS, Bhaumik R, Ren X. Innate immunity in the postmortem brain of depressed and suicide subjects: role of Toll-like receptors. Brain Behav Immun. 2019;75:101–11.

112. Steiner J, Bielau H, Brisch R, Danos P, Ullrich O, Mawrin C, et al. Immunological aspects in the neurobiology of suicide: elevated microglial density in schizophrenia and depression is associated with suicide. J Psychiatr Res. 2008;42(2):151–7.
113. Torres-Platas SG, Cruceanu C, Chen GG, Turecki G, Mechawar N. Evidence for increased microglial priming and macrophage recruitment in the dorsal anterior cingulate white matter of depressed suicides. Brain Behav Immun. 2014;42:50–9.
114. Brisch R, Steiner J, Mawrin C, Krzyżanowska M, Jankowski Z, Gos T. Microglia in the dorsal raphe nucleus plays a potential role in both suicide facilitation and prevention in affective disorders. Eur Arch Psychiatry Clin Neurosci. 2017;267(5):403–15.
115. Busse M, Busse S, Myint AM, Gos T, Dobrowolny H, Müller UJ, et al. Decreased quinolinic acid in the hippocampus of depressive patients: evidence for local anti-inflammatory and neuroprotective responses? Eur Arch Psychiatry Clin Neurosci. 2015;265(4):321–9.
116. Torres-Platas SG, Hercher C, Davoli MA, Maussion G, Labonté B, Turecki G, et al. Astrocytic hypertrophy in anterior cingulate white matter of depressed suicides. Neuropsychopharmacology. 2011;36(13):2650–8.
117. O'Leary LA, Belliveau C, Davoli MA, Ma JC, Tanti A, Turecki G, et al. Widespread decrease of cerebral vimentin-immunoreactive astrocytes in depressed suicides. Front Psychiatry. 2021;12:640963.
118. Cobb JA, O'Neill K, Milner J, Mahajan GJ, Lawrence TJ, May WL, et al. Density of GFAP-immunoreactive astrocytes is decreased in left hippocampi in major depressive disorder. Neuroscience. 2016;316:209–20.
119. Köhler CA, Freitas TH, Stubbs B, Maes M, Solmi M, Veronese N, et al. Peripheral alterations in cytokine and chemokine levels after antidepressant drug treatment for major depressive disorder: systematic review and meta-analysis. Mol Neurobiol. 2018;55(5):4195–206.
120. Hannestad J, DellaGioia N, Bloch M. The effect of antidepressant medication treatment on serum levels of inflammatory cytokines: a meta-analysis. Neuropsychopharmacology. 2011;36(12):2452–9.
121. Hiles SA, Baker AL, de Malmanche T, Attia J. Interleukin-6, C-reactive protein and interleukin-10 after antidepressant treatment in people with depression: a meta-analysis. Psychol Med. 2012;42(10):2015–26.
122. Strawbridge R, Arnone D, Danese A, Papadopoulos A, Herane Vives A, Cleare AJ. Inflammation and clinical response to treatment in depression: a meta-analysis. Eur Neuropsychopharmacol. 2015;25(10):1532–43.
123. Wang L, Wang R, Liu L, Qiao D, Baldwin DS, Hou R. Effects of SSRIs on peripheral inflammatory markers in patients with major depressive disorder: a systematic review and meta-analysis. Brain Behav Immun. 2019;79:24–38.
124. Więdłocha M, Marcinowicz P, Krupa R, Janoska-Jaździk M, Janus M, Dębowska W, et al. Effect of antidepressant treatment on peripheral inflammation markers - a meta-analysis. Prog Neuropsychopharmacol Biol Psychiatry. 2018;80(Pt C):217–26.
125. Beurel E, Toups M, Nemeroff CB. The bidirectional relationship of depression and inflammation: double trouble. Neuron. 2020;107(2):234–56.
126. Syed SA, Beurel E, Loewenstein DA, Lowell JA, Craighead WE, Dunlop BW, et al. Defective inflammatory pathways in never-treated depressed patients are associated with poor treatment response. Neuron. 2018;99(5):914–24.e3.
127. Piletz JE, Halaris A, Iqbal O, Hoppensteadt D, Fareed J, Zhu H, et al. Pro-inflammatory biomakers in depression: treatment with venlafaxine. World J Biol Psychiatry. 2009;10(4): 313–23.
128. Lehtimäki K, Keränen T, Huuhka M, Palmio J, Hurme M, Leinonen E, et al. Increase in plasma proinflammatory cytokines after electroconvulsive therapy in patients with depressive disorder. J ECT. 2008;24(1):88–91.
129. Feeney A, Hock RS, Freeman MP, Flynn M, Hoeppner B, Iosifescu DV, et al. The effect of single administration of intravenous ketamine augmentation on suicidal ideation in

treatment-resistant unipolar depression: results from a randomized double-blind study. Eur Neuropsychopharmacol. 2021;49:122–32.
130. Wang SM, Kim NY, Na HR, Lim HK, Woo YS, Pae CU, et al. Rapid onset of intranasal esketamine in patients with treatment resistant depression and major depression with suicide ideation: a meta-analysis. Clin Psychopharmacol Neurosci. 2021;19(2):341–54.
131. Nowak W, Grendas LN, Sanmarco LM, Estecho IG, Arena ÁR, Eberhardt N, et al. Pro-inflammatory monocyte profile in patients with major depressive disorder and suicide behaviour and how ketamine induces anti-inflammatory M2 macrophages by NMDAR and mTOR. EBioMedicine. 2019;50:290–305.
132. Köhler-Forsberg O, Lydholm CN, Hjorthøj C, Nordentoft M, Mors O, Benros ME. Efficacy of anti-inflammatory treatment on major depressive disorder or depressive symptoms: meta-analysis of clinical trials. Acta Psychiatr Scand. 2019;139(5):404–19.
133. Lehrer S, Rheinstein PH. Nonsteroidal anti-inflammatory drugs (NSAIDs) reduce suicidal ideation and depression. Discov Med. 2019;28(154):205–12.
134. Kappelmann N, Lewis G, Dantzer R, Jones PB, Khandaker GM. Antidepressant activity of anti-cytokine treatment: a systematic review and meta-analysis of clinical trials of chronic inflammatory conditions. Mol Psychiatry. 2018;23(2):335–43.
135. Tyring S, Bagel J, Lynde C, Klekotka P, Thompson EH, Gandra SR, et al. Patient-reported outcomes in moderate-to-severe plaque psoriasis with scalp involvement: results from a randomized, double-blind, placebo-controlled study of etanercept. J Eur Acad Dermatol Venereol. 2013;27(1):125–8.
136. Tyring S, Gottlieb A, Papp K, Gordon K, Leonardi C, Wang A, et al. Etanercept and clinical outcomes, fatigue, and depression in psoriasis: double-blind placebo-controlled randomised phase III trial. Lancet. 2006;367(9504):29–35.
137. Loftus EV, Feagan BG, Colombel JF, Rubin DT, Wu EQ, Yu AP, et al. Effects of adalimumab maintenance therapy on health-related quality of life of patients with Crohn's disease: patient-reported outcomes of the CHARM trial. Am J Gastroenterol. 2008;103(12):3132–41.
138. Menter A, Augustin M, Signorovitch J, Yu AP, Wu EQ, Gupta SR, et al. The effect of adalimumab on reducing depression symptoms in patients with moderate to severe psoriasis: a randomized clinical trial. J Am Acad Dermatol. 2010;62(5):812–8.
139. Simpson EL, Gadkari A, Worm M, Soong W, Blauvelt A, Eckert L, et al. Dupilumab therapy provides clinically meaningful improvement in patient-reported outcomes (PROs): a phase IIb, randomized, placebo-controlled, clinical trial in adult patients with moderate to severe atopic dermatitis (AD). J Am Acad Dermatol. 2016;75(3):506–15.
140. Langley RG, Feldman SR, Han C, Schenkel B, Szapary P, Hsu MC, et al. Ustekinumab significantly improves symptoms of anxiety, depression, and skin-related quality of life in patients with moderate-to-severe psoriasis: results from a randomized, double-blind, placebo-controlled phase III trial. J Am Acad Dermatol. 2010;63(3):457–65.
141. Sun Y, Wang D, Salvadore G, Hsu B, Curran M, Casper C, et al. The effects of interleukin-6 neutralizing antibodies on symptoms of depressed mood and anhedonia in patients with rheumatoid arthritis and multicentric Castleman's disease. Brain Behav Immun. 2017;66:156–64.
142. Raison CL, Rutherford RE, Woolwine BJ, Shuo C, Schettler P, Drake DF, et al. A randomized controlled trial of the tumor necrosis factor antagonist infliximab for treatment-resistant depression: the role of baseline inflammatory biomarkers. JAMA Psychiatry. 2013;70(1):31–41.
143. McIntyre RS, Subramaniapillai M, Lee Y, Pan Z, Carmona NE, Shekotikhina M, et al. Efficacy of adjunctive infliximab vs placebo in the treatment of adults with bipolar I/II depression: a randomized clinical trial. JAMA Psychiatry. 2019;76(8):783–90.
144. Mansur RB, Delgado-Peraza F, Subramaniapillai M, Lee Y, Iacobucci M, Rodrigues N, et al. Extracellular vesicle biomarkers reveal inhibition of neuroinflammation by infliximab in association with antidepressant response in adults with bipolar depression. Cells. 2020;9(4):895.

145. Services UDoHH. Child maltreatment; 2012.
146. McCloskey LA, Walker M. Posttraumatic stress in children exposed to family violence and single-event trauma. J Am Acad Child Adolesc Psychiatry. 2000;39(1):108–15.
147. Costello EJ, Angold A. Developmental psychopathology and public health: past, present, and future. Dev Psychopathol. 2000;12(4):599–618.
148. Nemeroff CB. Paradise lost: the neurobiological and clinical consequences of child abuse and neglect. Neuron. 2016;89(5):892–909.
149. Felitti VJ, Anda RF, Nordenberg D, Williamson DF, Spitz AM, Edwards V, et al. Relationship of childhood abuse and household dysfunction to many of the leading causes of death in adults. The Adverse Childhood Experiences (ACE) Study. Am J Prev Med. 1998;14(4): 245–58.
150. Dube SR, Anda RF, Felitti VJ, Chapman DP, Williamson DF, Giles WH. Childhood abuse, household dysfunction, and the risk of attempted suicide throughout the life span: findings from the Adverse Childhood Experiences Study. JAMA. 2001;286(24):3089–96.
151. Fergusson DM, Woodward LJ, Horwood LJ. Risk factors and life processes associated with the onset of suicidal behaviour during adolescence and early adulthood. Psychol Med. 2000;30 (1):23–39.
152. Afifi TO, Enns MW, Cox BJ, Asmundson GJ, Stein MB, Sareen J. Population attributable fractions of psychiatric disorders and suicide ideation and attempts associated with adverse childhood experiences. Am J Public Health. 2008;98(5):946–52.
153. McCauley J, Kern DE, Kolodner K, Dill L, Schroeder AF, DeChant HK, et al. Clinical characteristics of women with a history of childhood abuse: unhealed wounds. JAMA. 1997;277(17):1362–8.
154. Brezo J, Paris J, Tremblay R, Vitaro F, Hébert M, Turecki G. Identifying correlates of suicide attempts in suicidal ideators: a population-based study. Psychol Med. 2007;37(11):1551–62.
155. Isohookana R, Riala K, Hakko H, Räsänen P. Adverse childhood experiences and suicidal behavior of adolescent psychiatric inpatients. Eur Child Adolesc Psychiatry. 2013;22(1): 13–22.
156. Lin D, Li X, Fan X, Fang X. Child sexual abuse and its relationship with health risk behaviors among rural children and adolescents in Hunan, China. Child Abuse Negl. 2011;35(9):680–7.
157. Chen LP, Murad MH, Paras ML, Colbenson KM, Sattler AL, Goranson EN, et al. Sexual abuse and lifetime diagnosis of psychiatric disorders: systematic review and meta-analysis. Mayo Clin Proc. 2010;85(7):618–29.
158. Copeland WE, Wolke D, Angold A, Costello EJ. Adult psychiatric outcomes of bullying and being bullied by peers in childhood and adolescence. JAMA Psychiatry. 2013;70(4):419–26.
159. Brezo J, Paris J, Vitaro F, Hébert M, Tremblay RE, Turecki G. Predicting suicide attempts in young adults with histories of childhood abuse. Br J Psychiatry. 2008;193(2):134–9.
160. Turecki G. The molecular bases of the suicidal brain. Nat Rev Neurosci. 2014;15(12):802–16.
161. Dube SR, Fairweather D, Pearson WS, Felitti VJ, Anda RF, Croft JB. Cumulative childhood stress and autoimmune diseases in adults. Psychosom Med. 2009;71(2):243–50.
162. Paras ML, Murad MH, Chen LP, Goranson EN, Sattler AL, Colbenson KM, et al. Sexual abuse and lifetime diagnosis of somatic disorders: a systematic review and meta-analysis. JAMA. 2009;302(5):550–61.
163. Leserman J, Drossman DA, Li Z, Toomey TC, Nachman G, Glogau L. Sexual and physical abuse history in gastroenterology practice: how types of abuse impact health status. Psychosom Med. 1996;58(1):4–15.
164. Dong M, Giles WH, Felitti VJ, Dube SR, Williams JE, Chapman DP, et al. Insights into causal pathways for ischemic heart disease: adverse childhood experiences study. Circulation. 2004;110(13):1761–6.
165. Padgett DA, Glaser R. How stress influences the immune response. Trends Immunol. 2003;24 (8):444–8.
166. Webster JI, Tonelli L, Sternberg EM. Neuroendocrine regulation of immunity. Annu Rev Immunol. 2002;20:125–63.

167. Shanks N, Lightman SL. The maternal-neonatal neuro-immune interface: are there long-term implications for inflammatory or stress-related disease? J Clin Invest. 2001;108(11):1567–73.
168. Pace TW, Mletzko TC, Alagbe O, Musselman DL, Nemeroff CB, Miller AH, et al. Increased stress-induced inflammatory responses in male patients with major depression and increased early life stress. Am J Psychiatry. 2006;163(9):1630–3.
169. Danese A, Pariante CM, Caspi A, Taylor A, Poulton R. Childhood maltreatment predicts adult inflammation in a life-course study. Proc Natl Acad Sci U S A. 2007;104(4):1319–24.
170. Danese A, Moffitt TE, Pariante CM, Ambler A, Poulton R, Caspi A. Elevated inflammation levels in depressed adults with a history of childhood maltreatment. Arch Gen Psychiatry. 2008;65(4):409–15.
171. Crosswell AD, Bower JE, Ganz PA. Childhood adversity and inflammation in breast cancer survivors. Psychosom Med. 2014;76(3):208–14.
172. Slopen N, Kubzansky LD, McLaughlin KA, Koenen KC. Childhood adversity and inflammatory processes in youth: a prospective study. Psychoneuroendocrinology. 2013;38(2):188–200.

# Part III

# Inflammation and Therapeutic Interventions

# Effects of Current Psychotropic Drugs on Inflammation and Immune System

18

Shvetank Bhatt, Arghya Kusum Dhar, Malay Kumar Samanta, and Ashish Suttee

**Abstract**

The immune system and inflammation are involved in the pathological progression of various psychiatric disorders such as depression or major depressive disorder (MDD), generalized anxiety disorder (GAD) or anxiety, schizophrenia, Alzheimer's disease (AD), and Huntington's disease. It is observed that levels of inflammatory cytokines such as tumor necrosis factor-α (TNF-α), interleukin-1β (IL-1β), and other markers are highly increased in the abovementioned disorders. The inflammation and immune component also lead to enhance the oxidative stress. The oxidative stress and increased production of reactive oxygen species (ROS) are considered as important factors that are involved in pathological progression of psychiatric disorders. Increase production of ROS is associated with excessive inflammation followed by cell necrosis and death. The psychotropic drugs are mainly work through modulations of neurotransmitter system. However, it is evident that inflammation and immune modulation are also having important role in the progression of psychiatric disorders. Rationale of the use of current psychotropic drugs is modulation of immune system by them. However, the effects of psychotropic drugs on the immune system and how these might contribute to their efficacy remain largely unclear. The drugs may act through

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modification of inflammation and related markers. The main purpose of this book chapter is to address the role of current psychotropic drugs on inflammation and immune system. Moreover, it will also address the role of inflammation in the progression of psychiatric disorders.

**Keywords**

Inflammation · Immune system · Oxidative stress · Psychotropic drugs · TNF-α

## 18.1 Introduction

The psychotropic medications can be divided in five main subtypes, namely antidepressants, antianxiety drugs, CNS stimulants, antipsychotics, and mood stabilizers [1]. There are changes in neurotransmitter levels that have been observed in these disorders, and the drugs mainly work through the modification in the levels of various neurotransmitters (NTs) such as dopamine (DA), serotonin or 5-hydroxytryptamine (5-HT), norepinephrine (NE), glutamate, gamma-aminobutyric acid (GABA), acetylcholine (Ach), etc. [2]. The abovementioned neuropsychiatric disorders and inflammation are closely associated with each other in a bidirectional loop. Neuropsychiatric disorders facilitate inflammatory reactions on the other way inflammation promotes depression and other neuropsychiatric disorders. The patients affected with the psychiatric disorders have cardinal feature of inflammation including amplified circulating levels of inflammatory inducers, activated sensors, and inflammatory mediators targeting all tissues [3]. Mood behavior and cognition can be affected by the pro-inflammatory cytokines. These cytokines are associated with reduction in brain monoamine levels, activating neuroendocrine responses, promoting excitotoxicity (increased glutamate levels), and impairing synaptic plasticity. Increasing evidence suggests that modifications in regulation of neuroendocrine system, biotransformation, brain–gut–microbiome axis, oxidative stress, the use of prebiotics–probiotics, and negative health behaviors are important triggers of inflammation [4, 5]. Finally, recent data indicate that early-life stress is linked with overt inflammation prior to the development of neuropsychiatric disorders. Various clinical studies also indicated the crucial role of immune system in the progression of psychiatric diseases, while basic biology has revealed that the brain has an active and multicellular resident immune system that coordinates with peripheral immunity and affects behavior [6]. Neuroimmunology may act as an important perspective in the progression of psychiatric disorders.

## 18.2 Pathophysiology of Different Psychiatric Disorders

### 18.2.1 Depression or Major Depressive Disorder (MDD)

Latest report of WHO states that worldwide 264 million people are affected with the mental disorders. MDD is a state of prolonged sadness [7]. As per the monoamine hypothesis, mainly three NTs, namely, 5-HT, NE, and DA, are involved in depression. The levels of above three NTs decreased in the brain of depressed person [8, 9]. Glutamate and GABA are the other NTs that are involved in the pathophysiology of depression. GABA is an inhibitory NT of CNS, and glutamate is an excitatory NT. Current studies suggested that these NTs are involved in neuronal plasticity, long-term potentiation (LTP), limbic system development, and modification in frontal cortex and hippocampus volume [10] [11]. It is well recognized now that mutations in certain genes are also involved in the progression of depression. Genes like SLC6A4 (previously known as SERT), DRDR4, SLC6A4 or 5-HTT, and TPH2 are associated with pathological progression of MDD [12]. In addition, dysfunction of hypothalamic-pituitary-adrenal (HPA) axis, amplified oxidative stress, and excessive ROS production is also associated with pathophysiology of depression. Oxidative stress can be correlated with inflammation. Increase oxidative stress leads to increase in the levels of various pro-inflammatory peripheral biomarkers. Increased activity of C-reactive protein (CRP), TNF-α, and interferon-α has been observed in depressed patients [13, 14].

### 18.2.2 Anxiety or Generalized Anxiety Disorder (GAD)

Anxiety is a condition of fear and apprehension as important component. According to data from WHO, an estimated 3.6% of the population globally had anxiety is 264 million people [15]. More than 50% percent patients affected with depression have comorbidity of anxiety. According to the US National Comorbidity Survey, the prevalence of co-occurrence with other psychiatric disorders and any type of anxiety disorder in MDD patients is 76.7% and 56.8%, respectively [16]. Epidemiological data also indicate that 59% of subjects with anxiety disorder satisfy the criteria of depression [17]. The etiologic factors of anxiety include stress, comorbidity with other disorders such as diabetes and depression, genetic, first-degree relatives with generalized anxiety disorder (25%), and environmental factors like child abuse and substance abuse. The major NTs are involved in the anxiety are 5-HT, NE, and GABA. Lower activity of serotonergic system and elevated noradrenergic system activity are responsible for the pathophysiological progression of anxiety [18, 19]. The disorder is highly comorbid with MDD. The involvement of increased oxidative stress and dysregulation of HPA axis is common in the progression of anxietyand MDD. Increased ROS production and abnormality in the negative feedback mechanism of HPA axis is also observed in anxiety as seen with MDD [20]. The involvements of immune component with psychological disorders are studied in "Psychoneuroimmunology" [21]. The disorder anxiety has also

involvement of immune and inflammatory pathways in the pathological progression of the disease. The psychological stress is reported to affect the production of cytokines, proposing potential significance of this mediator to the psychological health. Moreover, signaling of cytokine in the brain is known to modulate the crucial brain functions including metabolism of neurotransmitters, physiology of neuroendocrine system, synaptic plasticity, and the neural circuitry of mood [22].

The main treatment of anxiety includes the use of selective serotonin reuptake inhibitors (SSRI), benzodiazepines, and psychotherapy [23]. Among psychotherapeutic treatments, cognitive behavior therapies have been extensively used and have wide evidence. Benzodiazepines such as diazepam, alprazolam, nitrazepam, etc. are effective in reducing anxiety symptoms by modulation of GABA NT-mediated signal transduction mechanism, but their use is restricted by the risk of abuse and adverse event profiles [24]. In addition to above treatments, SSRIs are also useful in the treatment of anxiety and depression. Moreover, serotonin-norepinephrine reuptake inhibitors (SNRI) are also used frequently as a first-line treatment for anxiety disorders [25].

### 18.2.3 Alzheimer's Disease (AD)

AD was first identified by Alois Alzheimer in a 51-year-old female patient with memory loss, confusion, and other psychiatric symptoms in 1906 [26]. In the same patient's brain, Alzheimer found plaques, neurofibrillary tangles, astrogliosis, and neuronal death [27]. AD is now the most common neurological condition. AD causes dementia, cognitive decline, and neuronal death. Globally, 131.5 million people will have dementia by 2050 [28].

AD causes broad brain atrophy due to severe neuronal degeneration and synapse loss in the hippocampus and cortex [29]. The key hallmarks of AD were thought to be amyloid beta (Aβ) peptide accumulation and neurofibrillary tangles (NFT). However, neuroinflammation has recently emerged as a third disease hallmark [30].

It is the abnormal presence of Aβ plaques and neurofibrillary tangles in the brain that defines AD. Although the causes and course of AD are unknown, postmortem diagnosis is achievable. The amyloid cascade hypothesis (ACH) blames Aβ build-up for AD etiology. The Aβ protein comes from the amyloid precursor protein (APP), which is prevalent in CNS cells and required for normal brain development and adult neural plasticity [31, 32]. APP can be processed in two ways: amyloidogenic and non-amyloidogenic [32].

Incorrect cleavage of the APP results in Aβ monomers that combine to form oligomeric Aβ and eventually fibrils and plaques. Understanding the mechanics of Aβ monomer formation, clearance, and aggregation into oligomeric Aβ is critical to understanding AD pathogenesis. APP is normally processed via non-amyloidogenic pathway involving proteolysis by α- and λ-secretases, resulting in soluble fragments [32, 33]. While Aβ is soluble, it has the potential to form oligomers, which then aggregate to create the amyloid plaques associated with AD [32]. Amyloidogenic processing of APP involves an initial cleavage by β-secretase 1 enzyme (BACE1)

followed by a second three step cleavage by the γ-secretase enzyme to generate insoluble Aβ, which aggregates to build β-amyloid plaques in the brain [34].

Even if the specific mechanism is unknown, genetic variants that influence A secretion or plaque development have been shown to affect AD risk. Familial Alzheimer's disease (FAD) is caused by mutations in the APP or presenilins 1 and 2, which code for the enzymes involved in Aβ cleavage. FAD is genetic. Aβ plaque load found in postmortem brains of elderly persons without dementia has no association to their level of cognitive impairment [32, 35]. Aβ plaques can grow up to 10 years before symptoms or diagnosis; therefore, their role in AD etiology is uncertain. An accumulation of neurofibrillary tangles (NFTs) is another feature of AD. Hyperphosphorylation of Tau protein forms NFTs, which are required for microtubule stability [32].

Tau must be phosphorylated before it can be carried by microtubules. Then dephosphorylation restores Tau to the microtubule. Multiple Tau phosphorylation in AD promotes microtubule collapse and disruption of several cellular activities, from protein trafficking to overall cellular structure [36–38]. pTau generates paired helical fragments that eventually form neurofibrillary tangles [39–43]. Neurons begin to misfire and finally die because of pTau tangle accumulation and loss of cellular function [36].

Aβ plaque accumulation leads to neurofibrillary tangles, which lead to neuritic injury and cell death [44–46]. They tend to be more detrimental than plaques in terms of disease severity and cognitive decline [32, 47, 48]. Suppressing long-term potentiation in APP mutant mice prior to plaque formation has been found to affect synaptic function, calcium homeostasis, neuroinflammation, and oxidative stress. Most ACH-based AD treatments target Aβ synthesis. While solanezumab and aducanumab showed early promise, they failed in phase 3 clinical trials [32, 49, 50]. Two prominent occurrences show the need for disease models beyond ACH. On autopsy and imaging studies, NFTs are found in elderly people [51]. Individual reactions to Aβ deposits and NFTs may influence AD susceptibility. It reduced Aβ burden in both human and animal models but did not affect disease progression. Some patients got better, but AD continued. Aβ may cause an inflammatory reaction to Aβ and NFT. Neuroinflammation is increasingly recognized as a major etiology of AD [32, 52].

New AD features may help explain disease development and link the other two major disorders. In addition to Aβ plaques and NFT, AD patients show chronic brain inflammation [53]. Inflammation has been observed in postmortem AD tissues and preclinical AD model systems [53]. Disruption of anti-inflammatory/pro-inflammatory signal balance causes chronic inflammation (AD) (neuroinflammation). Chronic neuroinflammation is caused by microglia and cytokines. Neuroimmunological responses are not unique to AD. According to some research, people with Parkinson's disease, chronic traumatic encephalopathy (CTE), amyotrophic lateral sclerosis (ALS), and multiple sclerosis (MS) have higher signs of inflammation in their brains. A long-term immune response is now a hallmark of neurodegenerative diseases [53].

The extended inflammatory response in AD patients' brains was originally thought to be a consequence to neuronal loss. Recent research demonstrates that prolonged immune responses in the brain produce neurodegeneration and exacerbate Aβ and NFT disorders. Inflammation may be a link between early Aβ pathophysiology and later NFT formation [53–56].

### 18.2.4 Schizophrenia

Schizophrenia is a serious mental condition that affects around 1% of the world's population. Early childhood interactions between genetic predisposition and environmental stresses and subsequent molecular neurodegeneration play a role in the development of schizophrenia. Positive symptoms like hallucinations, delusions, and disorganized speech are common in schizophrenia, as are negative symptoms like social disengagement, lack of desire, and cognitive symptoms including attention and learning difficulties [57, 58].

Schizophrenia is assumed to be caused by a neurodevelopmental disorder. Defective connection between dorsolateral prefrontal cortex and subcortical regions is a contributing factor to the development of autism. The cause and pathophysiology of schizophrenia are still unknown, but there is some evidence that suggest dopamine, serotonin, gamma-aminobutyric acid (GABA), and glutamate system alterations have a role in the onset of schizophrenia symptoms [57].

Schizophrenia has been linked to dopaminergic system abnormalities ever since the dopamine hypothesis was first proposed [59, 60]. Schizophrenia patients have altered dopaminergic transmission in the mesocortex and mesolimbic ganglia. The mesolimbic dopaminergic neurotransmission, on the other hand, is overactive in schizophrenia, even though the mesocortical dopaminergic transmission to the prefrontal cortex is hypoactive [57].

Dopamine 2 (D2) receptor stimulation in the subcortical regions is assumed to be responsible for schizophrenia's positive symptoms, while dopamine 1 (D1) receptor stimulation in the cortex is responsible for schizophrenia's negative symptoms. Current antipsychotic drugs (APDs), which block D2 receptors to diminish mesolimbic dopamine transmission, affect clinical remission in schizophrenia patients [61].

While the dopamine hypothesis still dominates translational research in schizophrenia, new data reveals that abnormal immunological pathways in the peripheral and central nervous systems influence the etiology and pathophysiology of symptoms [57].

Current research suggests that neuroinflammation may play a role in the development of schizophrenia. If microglial activation is uncontrolled and pro-inflammatory cytokines are released into the brain, schizophrenia can result. By inducing microglial activation, pro-inflammatory cytokines in the central nervous system trigger inflammation, which in turn leads to neurodegeneration (Fig. 18.2) [57].

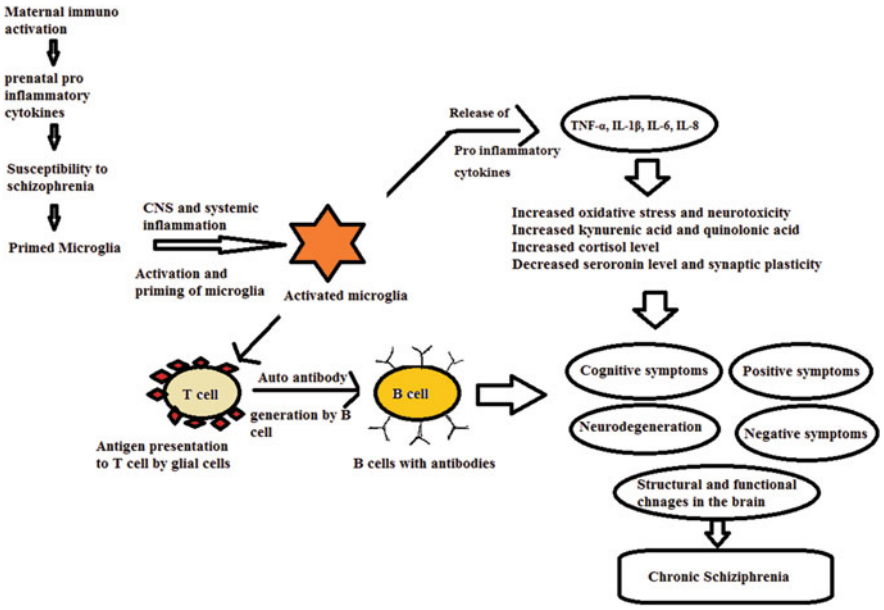


**Fig. 18.2** Depicts progression of schizophrenia. Various pro-inflammatory cytokines are responsible for the activation of microglia. The activated microglia releases pro-inflammatory cytokines, abnormal oxidative stress, increased kynurenic and quinolinic acids, increased cortisol level, decreased serotonin level, synaptic plasticity, and present antigen to T cells and leads to generation of autoantibody by B cells. The overall action leads to generation of positive and negative symptoms of psychoses and neuronal degeneration

#### 18.2.4.1 Neurodevelopmental Approach and Infection Link to Schizophrenia

Psychiatric disorders like schizophrenia stem from brain development. Schizophrenia is caused by a mix of environmental, genetic, and developmental factors [62]. In schizophrenia, brain damage begins early in life and takes a long time to manifest [62].

Some researchers believe schizophrenia increases the risk of developing other CNS disorders, such as autism, Parkinson's disease, AD, and MS. This may be due to the fetus being exposed to inflammatory modulators during development, causing brain disruptions or malfunctions [63, 64]. Pregnant women who are exposed to viral or bacterial pathogens are more prone to schizophrenia. A higher risk of schizophrenia in offspring has been linked to maternal infections like toxoplasmosis and rubella [57].

Several epidemiological studies have linked prenatal infection to increased risk of schizophrenia in children [59]. These findings support the theory that maternal immune activation (MIA) increases the risk of schizophrenia by altering the fetal brain's development [65].

Schizophrenia-like symptoms, such as a disturbed pre-pulse inhibition in the offspring, have been observed in animal models of schizophrenia that were

stimulated during pregnancy by viral or bacterial agents (such as influenza) [65]. Prenatal or perinatal exposure to infections has been shown to be a risk factor for schizophrenia in both animal models and humans [65].

Interleukin (IL)-1, IL-6, and TNF-a are thought to contribute to MIA's detrimental effects on the fetal brain [65]. Endotoxins like lipopolysaccharide and polyinosinic/polycytidylic acid were found to increase the expression of pro-inflammatory cytokines (polyI/C) in rat fetal brain. The inflammatory cytokine IL-6 has been linked to structural and functional deficits in the developing fetal brain [65].

Infections have also been linked to schizophrenic symptoms in human studies [66]. Infections of the respiratory, genital, and reproductive tracts increased the risk of schizophrenia in children [66]. Prenatal *Toxoplasma gondii* infection has been linked to schizophrenia [66]. Encroachments on the CNS (especially prenatal infections) raise the risk of later schizophrenia [66]. For decades, schizophrenics have been tested for viral antibody titers. The results may be inconsistent because interfering factors such as antibody levels associated with medication state were not controlled [66]. The "infectious index" of schizophrenia patients was found to be higher than that of healthy controls [66].

Interleukin-8 (IL-8) is a cytokine that has been linked to an increased risk of schizophrenia in children [66]. Lower volumes of the right posterior cingulate and left entorhinal cortex and increased volumes of the ventricles were linked to increased maternal IL-8 levels during pregnancy [66].

Childhood autoimmune diseases are linked to adolescent and adult psychosis [67]. First-degree relatives of schizophrenics are more prone to autoimmune diseases [67]. In people with autoimmune disease, the risk of schizophrenia increases linearly with the number of severe infections [67]. As a result, the links between schizophrenia and infections and autoimmune diseases suggest an inflammatory immune response [67]. Along with its own effects on the brain, inflammation is thought to increase the blood–brain barrier permeability and aid in immune component penetration [67].

Schizophrenia has been linked to abnormal ROS and inflammation. Stress may contribute to the pathogenesis of schizophrenia by increasing pro-inflammatory cytokines and even contributing to the development of a long-term pro-inflammatory state. Immune system changes affect glutamate and dopaminergic neurotransmission. The neuroactive metabolite indoleamine 2,3-dioxygenase influences serotonergic and glutamatergic neurotransmissions [57].

There is evidence of increased density and activation of microglia and brain-resident immune cells, throughout the illness [57]. However, recent research has shown that the immune system, systemic inflammation, and the brain are all intertwined and can lead to changes in behavior, mood, and perception [57]. Understanding the neuroinflammatory mechanisms involved in schizophrenia may help find potential therapeutic targets to help reduce mortality and morbidity [57].

## 18.3 Trauma- and Stressor-Related Disorders

Trauma and stress are responsible for traumatic and stressful events. The two major trauma-related disorders are acute stress disorder and posttraumatic stress disorder (PTSD). Accumulating evidence demonstrated that excess of inflammation plays a crucial role in associationship between stress and stress-related diseases. Trauma and stress are also associated with substance abuse to a great extent [68]. It is well accepted now that stress also weakens the immune system. The adrenergic system is stimulated in PTSD. In addition, disruption of HPA axis and negative feedback mechanism is also observed in PTSD. The traumatic brain injury model in rats demonstrated the hyper-agitated behavior post injury. The animals showed increased activity in open field test and phase aversion and aggressive behavior in elevated plus maze test. In addition, the animals also showed the anhedonia behavior [69]. Cognitive behavior therapy (CBT) is considered as first-line treatment for stress and trauma disorders. However, benzodiazepines can be used to get relieved from agitation and sleep disturbances. The hallmark events during trauma include Wallerian degeneration of axons, dysfunction of mitochondria, neuronal excitotoxicity, oxidative stress, and cell death due to destructions of neurons and glial cells [70]. Many disturbing changes are taken place in the neurotransmitter levels after the injury or trauma. The modification in neurotransmitters is not specific and depends on the area of injury.

### 18.3.1 Associationship of Depression with Inflammation and Immune System

It is well known that inflammatory processes are having influence on the progression of depression. Dysregulation of both innate and adaptive immune response has been implicated in the pathophysiology of depression. The immune component hinders with favorable prognosis in addition to antidepressant treatment. Presence of inflammation may influence the susceptibility to depression [71]. In our brain, microglia and other CNS cells have crucial role in CNS functions like neuroplasticity. Excess and long-term inflammatory cytokine activity produces impairment of neurotransmitter signaling disruption of the synthesis, reuptake, and release of neurotransmitter. The brain has specialized cells known as glial cell namely microglia. It is composed of 5–10% of total brain cells and carries out functions similar to macrophages, i.e., engulfment of external debris [72]. Recent evidence suggested that microglia cells are important for neurogenesis and synapse pruning. Microglia cells are activated in various neurodegenerative and neuropsychiatric diseases, where they have role in neuroinflammation [73]. However, more studies are required to get proper insight on the role of microglia in the progression of depression and other brain functions and disorders.

Controlling inflammation might have a useful therapeutic approach for the treatment of depression. Moreover, inflammation component has also linked with oxidative stress. Consequent to an increased OS, stimulation of pro-inflammatory

signaling pathways also associated with progression of depression. Depression is linked with an imbalance of factors associated with neurodegeneration and neuroprotection including brain-derived neurotrophic factor (BDNF) and NF-κB [74, 75]. MDD is also associated with various inflammatory processes such as enhancement of activity of pro-inflammatory cytokines, decreased nerve growth, and subsequent neuro-progression. Production of inflammatory markers IL-1 and IL-18, the formation of cellular pores in membrane, and leakage of the substances from the cells lead to cell death [76–78]. The subjects affected with depression have shown higher levels of IL-1, IL-6, TNF-α, and C-reactive protein (CRP) compared to nondepressed individuals [79, 80]. Inflammation is a component of innate immune system's response toward any injury or infection. The chief mediators of the inflammatory response, pro-inflammatory cytokines, such as interleukin (IL)-1β, interleukin (IL)-1 receptor antagonist (RA), interleukin (IL)- 6, tumor necrosis factor (TNF)-α, and interferon (IFN)-γ, have been recently demonstrated to associate with the brain and influence neuronal signaling, activity of neuroendocrine system, and structure and physiology of brain, thereby inducing the changes in the emotional, cognitive, and behavioral setup [3, 81]. Increase in the levels of pro-inflammatory cytokines can enhance pathological progression of MDD. The positive relationship is existing between inflammation and depression as suggested by various [80, 82].

Acute stress also leads to increase inflammation and further leads to behavioral and cognitive decline [83, 84]. Exposure to long-term stress causes dysfunction of endocrine and immune system functions that results in sustained low-grade inflammation, which involved in MDD pathogenesis [85]. Individuals exposed to early-life adversity (ELA) exhibit pronounced increased in the levels of pro-inflammatory cytokines [86].

### 18.3.2 Association of Anxiety with Inflammation and Immune System

Anxiety is a condition of fear and apprehension with insomnia. Most of the patients of depression also have comorbid anxiety. Anxiety may be present in various forms like panic disorder, social phobia, stress related, and obsessions and compulsions associated. Large evidence indicate that stress have ability to activate inflammatory pathways in the brain and peripherally [85, 87]. The communication exists between the neuroendocrine and immune systems [88]. Long-term stress activates HPA axis. This activation leads to higher levels of the glucocorticoids (GCs) such as cortisol in the blood. These GCs are well recognized for their anti-inflammatory and immunosuppressive activity. GCs reduce the expression of several pro-inflammatory cytokines (e.g., tumor necrosis factor α (TNF-α) and interleukin-6 (IL-6)) and enhance the expression of anti-inflammatory cytokines (e.g., IL-10 and TNF-β) [89]. However, recent studies by researchers have showed that GCs have pro-inflammatory potential on immune system. In addition, GCs also increase the expression of inflammasome NLRP3 and enhance the release of IL-1β in response to ATP. Cytoplasmic multi-protein senses exogenous and endogenous danger

signals and cleaves pro-inflammatory cytokines into mature cytokines such as IL-1β and IL-18 [90]. According to a case-controlled study, significant increase in the serum levels of IL-10, TNF-α, and IFN-γ has been observed in patients affected with GAD [91]. The above study investigated the balance of pro- and anti-inflammatory cytokines in the progression of GAD. The study demonstrated relatively increased pro-inflammatory response and decreased anti-inflammatory response.

### 18.3.3 Association of AD with Inflammation and Immune System

Neuroinflammation is linked to AD etiology because it is significantly expressed near A plaques and NFTs [92]. Amyloid plaque clearance and antioxidant mechanisms against ROS production are particularly effective in early AD [93]. Anti-inflammatory immune system cellular mediators are overproduced when oxidative stress increases in AD, increasing brain inflammation [94].

The BBB is a strict gatekeeper for systemic stimuli, including inflammatory signals. Neuroinflammation is triggered by stress, disease, and traumatic brain injury [95]. Inflammation has a favorable initial reaction, but when it persists, it causes neuronal dysfunction, damage, and loss [96]. Microglia are resident macrophages and perform basic immunological monitoring, and astrocytes have several functions and are essential for cell-to-cell communication [97].

The CNS is inflamed by microglia, astrocytes, and neurons. Microglial activation produces increased pro-inflammatory effects and neurotoxicity in AD. Inflammatory agents, oxidative stress, and neuroinflammation are all caused by activated microglia as demonstrated in Fig. 18.1 [98]. Inflammation by microglia releases neurotoxic cytokines. Microglia activation leads to neurotoxic changes that contribute to AD onset/progression. Astrocytes may be involved in AD development [98]. Aβ plaques stimulate astrocytes, increasing cytokine (IL-1 or IL-6) and oxidative stress [98]. Synaptic connections are disrupted, resulting in neuronal injury [99, 100]. Ingestion of oligomers causes extracellular annular protofibrils, which enhance oxidative stress and neuron death. Neurons may be involved in neuroinflammation through increasing inflammatory molecule expression. Neurons promote inflammation by exacerbating local inflammation [98].

Neuroinflammatory biomarkers are linked to AD progression. In 31 mild AD patients, hippocampal shrinkage, cognitive profile, and neuroinflammation were statistically associated [32]. On the other hand, anti-IFN Ab treatment cured AD and cognitive impairment in mice produced by TH1 cells [32]. Neuroinflammation via IL-1, IL-6, and NO seem to increase Tau hyperphosphorylation. They increase BACE enzyme activity and mRNA levels, which cleaves APP to generate Aβ [101].

Systemic inflammation, obesity, and traumatic brain injury all increase the risk of AD [102]. Inflammation may prime microglia prior to AD onset, making them more prone to activation [102]. Pro-inflammatory cytokines and chemokines may be secreted after Aβ activation, causing neuronal hyperexcitability and synaptic dysfunction. Previously thought to be passive observers during neuroinflammation, recent research shows that neurons can actually create inflammatory mediators

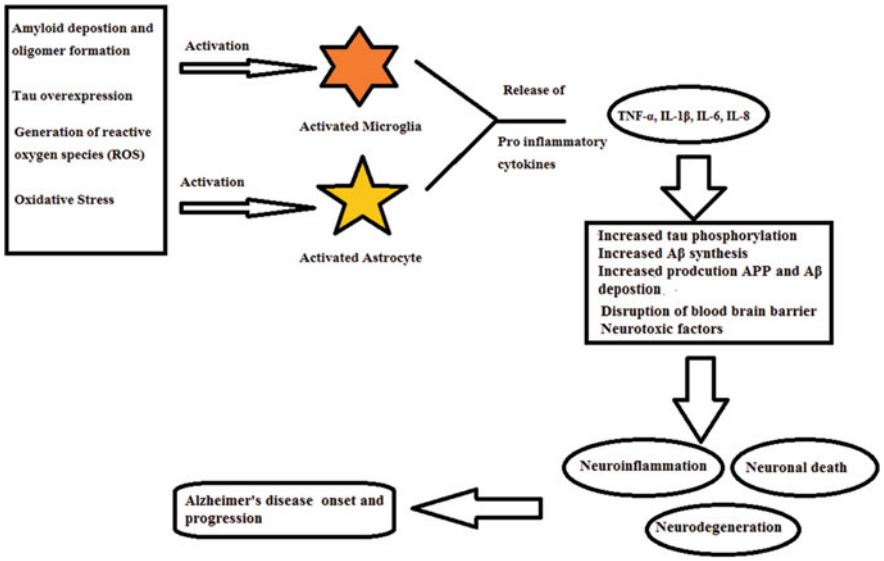


**Fig. 18.1** Depicts onset and progression of Alzheimer's disease. Abnormal Aβ deposition, hyperphosphorylation of Tau protein, and production of ROS lead to activation of microglia, and astrocytes lead to release of pro-inflammatory cytokines. Activation of cytokines results in neuroinflammation, neurodegeneration, and nerve cell death. IL: interleukin and TNF: tumor necrosis factor

[102]. Complement activation is critical in AD, because neurons manufacture the vast majority of the cascade's components. Pathogen-associated molecular patterns (PAMPS) and danger-associated molecular pattern (DAMPS) have the ability to activate the complement system. Complement can be activated by the buildup of extracellular Aβ or the discharge of components from dying neurons since C1q can directly bind to molecules like Aβ [102]. Additionally, AD patients have elevated levels of complement component synthesis in their neurons. COX-2-derived prostanoids and cytokines such IL-1β, IL-6, and tumor necrosis factor alpha (TNF-α) have also been linked to neurons [102].

Several investigations have indicated that in vivo LPS treatment increases $A\beta_{1-42}$ and decreases $A\beta_{1-40}$ [102, 103]. This finding links amyloidogenesis to neuroinflammation. However, the mechanisms of LPS-induced amyloidogenesis remain unexplained. Lee et al. found increased β- and γ-secretase activity in cortical and hippocampal regions of ICR mice and Sprague-Dawley rats treated with LPS, suggesting secretase activity is a contributing component [102, 103]. Pro-inflammatory cytokines including TNF and IL-1 have been demonstrated to increase β-secretase mRNA, protein, and enzyme activity. Lee et al. argue that LPS-induced inflammation affects APP processing by increasing β- and γ-secretase activity and thereby amyloidogenesis [102, 103]. Inflammatory mediators and cells involved with AD DAMP receptors on the surface of innate immune cells identify unfolded, misfolded, and aggregated proteins. In the realm of

neuroinflammation, aggregated Aβ functions as a DAMP, activating the innate immune system in the brain, resulting in pro-inflammatory cytokine production [104]. Following an initial buildup of extracellular oligomeric Aβ, microglia may be induced to produce an inflammatory response. As the pro-inflammatory response target self-DAMPS and pro-inflammatory cytokines damage neurons, a positive feedback loop is likely to form, resulting in disease development and chronic disease [102].

When it comes to peripheral innate immune responses, cytokines in the central nervous system (CNS) play an important role in controlling neuroinflammation. AD may cause neuronal damage in part because of the brain's pro-inflammatory environment. IL-1β and TNFα, for example, may have a direct impact on neuronal function [105]. Pro-inflammatory cytokines, notably TNF- α and IL-6, have been found in higher amounts in serum and brain tissue of AD patients than in controls [32, 106, 107]. The astrogliosis-inducing cytokine IL-1β was discovered in 30 times more glial cells, mainly microglia, in AD patient's tissue sections than in controls [108]. Patients with raised TNFα levels and lower cerebrospinal fluid (CSF) TGFβ concentrations are more likely to proceed from mild cognitive impairment to dementia. Microglia secreted pro-inflammatory and neurotoxic substances (IL-1β and iNOS) when stimulated by amyloid plaques in AD model rats [32].

Microglia and astrocytes are stimulated by cytokines to produce inducible nitric oxide synthase (iNOS), which is toxic to neurons at high doses [43]. iNOS has been found to be overexpressed in AD brains [102]. In vitro tests have confirmed this theory by demonstrating that microglia binding to Aβ results in the generation of reactive oxygen species (ROS) [102].

Study indicated that AD patients' microvasculature expressed higher levels of pro-inflammatory IL-1β, IL-6, TNF-α, and microvessel-associated monocyte chemoattractant protein than non-AD controls [109]. Immune cells physically connect with Aβ, in addition to pro-inflammatory chemical signatures. Several investigations have shown the presence of immune cells and proteins near Aβ plaques. The HLA-DR protein, which is typically produced by activated T cells, has been shown to colocalize with Aβ plaques in the cortex of AD patients, indicating that they may cross the BBB [110].

AD is substantially connected with ageing. Several studies have shown that healthy ageing impacts the brain's ability to trigger an immunological response [32]. Increased levels of pro-inflammatory cytokines like IL-1, IL-6, CD68, CD11b, and decreased levels of TLRs and anti-inflammatory cytokines like IL-10 and IL-4 have been associated to ageing [32]. Also, ageing has been associated to increased BBB permeability, allowing external influences, such as pro-inflammatory cytokines, to have a greater impact on brain homeostasis and neuroinflammatory processes [32].

### 18.3.4 Association of Schizophrenia with Inflammation and Immune System

When schizophrenia patients had acute psychotic exacerbations in the past, researchers found that the levels of pro-inflammatory cytokines (IL-1β., IL-6., and TNF-α.) in their peripheral blood were elevated, suggesting the presence of immunological alterations [111, 112]. Additionally, microglia, innate immune cells that reside in the CNS, secrete cytokines as a response to traumatic, infectious, and stressful events [113]. Immune system activation in the CNS, particularly through the activation of microglia during pregnancy, can be a "priming" mechanism as shown in Fig. 18.2 [66, 114–116].

Conditioned stimuli such as prior infections and environmental stress can increase the release of pro-inflammatory cytokines from primed microglia [117, 118]. During adolescence, the prefrontal cortex and the hippocampus are particularly vulnerable to external stressors, resulting in damage to these regions of the brain [119]. A previous study in mice found that IL-6 administration increased the sensitivity of mice to amphetamine [120, 121] and ketamine-induced [122] neurobiological insults, suggesting an intertwined relationship between pro-inflammatory cytokines and neurotransmitter systems. Irritation-induced brain damage can be caused in part by activated microglia that shift the kynurenine metabolism toward quinolinic acid (QA), which causes oxidative stress and neurotoxicity [123–125]. In animal studies, a high concentration of QA was found to disrupt the neurodevelopmental process and cause cognitive and behavioral alterations that are associated with schizophrenia [126, 127]. Although the role of kynurenine metabolites in schizophrenia has yet to be fully understood, sustained microglial activation may lead to further neurodegeneration and deterioration of the illness [128].

Previous studies have shown an increased density of microglia in postmortem brain analysis of patients with chronic schizophrenia, which supports the involvement of activated microglia [129, 130]. Patients with schizophrenia [131, 132] and those at high risk of psychosis [133] have activated microglia in their gray matter, according to in vivo neuroimaging studies using positron emission tomography.

The plasma cytokine levels of schizophrenia patients have been found to be elevated in many clinical studies. Prostaglandin E2, C-reactive protein, interleukin (IL)-1ß, IL-6, IL-8, and tumor necrosis factor (TNFα)-a serum/plasma levels are all indicators of an increased immune response in the peripheral tissue [111, 134].

The upregulation of peripheral IL-6, IL-1ß, TNF-α, soluble IL-1β antagonist, soluble IL-2 receptor, and IL-8 has been documented in studies performed in patients with first-episode schizophrenia receiving minimal treatment or no medication at all [135, 136]. IL-6, IL-12, TNF-α, IL-1ß, and interferon (IFN)-γ are elevated in the blood and cerebrospinal fluid of patients with acute relapse and the first episode of schizophrenia, according to a meta-analysis of previous research [26, 111].

The monocytic immune response, which produces and secretes pro-inflammatory cytokines, has also been found to be disturbed in schizophrenia

[136, 137]. Schizophrenia has been linked to an increase in the number of total white blood cells and monocytes in numerous studies [138].

The anti-inflammatory response increases in response to increased peripheral pro-inflammatory activity in schizophrenia [139]. This is supported by the rise in sIL-1RA and sIL-2R levels in the periphery [140]. To protect the organism from the harmful effects of the pro-inflammatory process, it is thought that the mentioned increase occurs as a result of the stimulation of the pro-inflammatory process [51, 111].

Chronic low-level inflammation [141] associated with immune sensitization may lead to a more severe disease process because of the abnormal expression of inflammatory genes in the monocyte/ macrophages in response to specific environmental factors, such as physical, physiological, or pathogen contact [51, 58].

Peripheral immune changes have been shown to influence brain functions and behaviors in a variety of neuropsychiatric conditions. At least 20–30% of patients have experienced acute psychotic episodes characterized by depression or excitation because of receiving repeated injections of purified and recombinant cytokines used to treat viral diseases and uncontrolled cancer [51, 142]. Pro-inflammatory cytokines may be produced in the brain as a result of peripheral immune stimulation, which may have an effect on the brain's functions. Increased expression of pro-inflammatory cytokines in mice's hypothalamus (IL-8, IL-1ß, and TNF-α) after intraperitoneal LPS injections has been documented and is linked to symptoms like decreased food intake [51, 143].

### 18.3.5 Association of Trauma- and Stressor-Related Disorders with Inflammation and Immune System

Considerable evidence suggested that the long-term stress is associated with inflammation. The level of CRP is increased due to chronic stress. Chronic stress also associated with glucocorticoid resistance including cytokine changes in response to stressful challenges. Within 24 h of TBI injury, dysfunction of BBB takes place. This dysfunction leads to infiltration of circulating neutrophils, monocytes, and lymphocytes into the injured parenchyma of brain [144]. Cerebrospinal fluid (CSF) analysis and postmortem tissue of TBI patients as well as tissue of TBI rodents demonstrated that polymononuclear leukocytes release complement factors and pro-inflammatory cytokines such as IL-1β, IL-6, and TNF-α, as exhibited by an increase in the corresponding mRNA and protein 24 h after the trauma [145] [146]. The sustained increase in the levels of cytokines is associated with modified permeability of BBB, edema formation, and neurological deficits. The TNF-α activation is associated with stimulation of apoptotic pathway. TNF-α interacts prominently with Fas ligand, which leads to activation of caspases that are essential for programmed cell death. In addition, chemokines like MIP-α, MCP-1, and IL-8 (CXCL8) are also predominantly upregulated after trauma, which further involved in the recruitment of leukocytes at the site of injury [147, 148]. In addition, upregulation of ICAM-1 and VCAM-1, which are ligands for endothelial and

leukocyte cell adhesion receptors, also governs the cross talk between WBC and immune cells with endothelium hence stimulating their recruitment at the site of injury [149]. Long-term inflammation is associated with recruitment of macrophages, activates micro glial cells, and promotes astrogliosis. Increased phagocytosis with continuous inflammatory response is demonstrated by macrophage accumulation and microglia activation in TBI survivors' years after injury [150].

## 18.4 Psychotropic Drugs

Current psychotropic drugs have been associated with modulation of immune system. However, the effect of psychotropic drugs on inflammation and immune system is still not clear. The available literature suggested that antidepressants like clomipramine and fluoxetine more consistently decreases the levels of pro-inflammatory cytokines such as IL-6, IFN-γ, and TNF-α while others like mirtazapine and venlafaxine tend to increase the levels of pro-inflammatory cytokines. Overall inflammatory markers need to hold important place in the discovery of psychotropic substances. The effect of these psychotropic drugs needs to be studied in detail [151].

### 18.4.1 Effect of Psychotropic Drugs on Inflammation

#### 18.4.1.1 Antidepressants on Inflammation

Increasing evidence of mediators of pro-inflammatory expression in major depression indicate that inflammatory mediators play a crucial role in the pathogenesis of depression [84]. If the above statement is true, the efficacy of antidepressants may be partially attributable to suppress the inflammation. The antidepressants can inhibit the levels of inflammatory mediators in addition to the modulation of neurotransmitters. The mechanism also includes the modification in the signaling of glial cells, which are considered as the main sources and targets of cytokines and other inflammatory mediators in the brain [152]. A pooled meta-analysis study conducted by Hannestad et al., 2011, also revealed that resolution of a depressive episodes may be associated with the normalization of the cytokine levels. However, the results are consistent with the possibility that inflammatory cytokines may contribute to depressive symptoms and that antidepressants block the effects of inflammatory cytokines on the brain [153]. In addition, the presence of inflammation in the patients of depression affects the outcome of first-line treatment for major depressive disorder. In this situation, immediately, one should be switched to the regimens that affect dopaminergic and glutaminergic transmission, or additionally, anti-inflammatory regimens can be added [154]. Moreover, a bidirectional relationship exists between depression and inflammation. Several studies have indicated the effect of anti-inflammatory drugs on depression. However, the results have been conflicting, and detrimental adverse effects may contraindicate the use of

anti-inflammatory agents. Several studies have reported antidepressant effects of anti-inflammatory treatment; however, the results have been conflicting, and detrimental adverse effects may contraindicate the use of anti-inflammatory agents.

#### 18.4.1.2 SSRIs, SNRIs, and Tricyclic Antidepressants on Inflammation

Accumulating evidence suggested that in in vitro studies SSRIs and SNRIs produces their effect vial modification in the signaling of inflammatory pathways. Ohgi et al., 2013, reported that SSRIs and SNRIs produce their effect via modification of the levels of pro-inflammatory cytokines TNFα and the anti-inflammatory cytokine, IL-10 in mice. According to study conducted by them, pre-treatment with SSRIs (fluoxetine and paroxetine), SNRIs (venlafaxine and duloxetine), or 5-hydroxytryptophan (5-HTP), a precursor of serotonin, attenuated LPS-induced increases in TNFα, whereas it increased serum levels of IL-10, in mice treated with LPS. The above evidence suggested that antidepressants also produce their effects in animal models via modification of inflammatory pathways. The above study also indicates the partial effect of serotonin in the signaling [155]. Moreover, Bhatt et al., 2017, also reported the effectiveness and potential of 5-$HT_3$ receptor antagonist, (4-benzylpiperazin-1-yl)(3-methoxyquinoxalin-2-yl) methanone (compound code: 6 g) in various animal models of depression and comorbid anxiety. The compound produces its action via modification of inflammatory pathway and showed its potential against depression comorbid with anxiety [156]. The in vitro studies with antidepressants demonstrated that drugs like clomipramine and fluoxetine, more consistently, reduces the levels of pro-inflammatory cytokines such as IL-6, IFN-γ, TNF-α; on the other hand, mirtazapine and venlafaxine are associated with increase in their levels. So, no clear-cut results are obtained with respect to effect of anti-depressants on inflammation [151].

#### 18.4.1.3 Tetracyclic Antidepressant: Mianserin

Mianserin is a tetracyclic antidepressant that has antihistaminic and hypno-sedative, but minimal anticholinergic potential. It is a weak norepinephrine reuptake inhibitor and predominantly stimulates the release of norepinephrine. It is also interacted with serotonergic receptors in central nervous system. The scientists have reported that structural modification in the antidepressant mianserin also leads to produce anti-inflammatory activity that may be independent of 5-hydroxytryptamine receptors. In the study, lead compound demonstrated a significant loss of serotonergic receptor binding. However, the compound retained the potential to inhibit endosomal toll-like receptor 8 (TLR-8) signaling primary human macrophages and spontaneous cytokine production from human rheumatoid synovial tissue equivalent to that previously observed for mianserin [157].

#### 18.4.1.4 Antianxiety Drugs and Posttraumatic Stress Disorder Medications on Inflammation

Anxiety is comorbid with depression. It is common in around 60–70% of the patients affected with depression. Anxiety comes when we think too much about the future. As discussed in the earlier sections of this chapter, pathophysiological progression of

anxiety and stress may be involved inflammation and immune component [158]. There are a good number of data available for the correlation of depression with inflammation as compared to anxiety with inflammation. Anxiety also comes hand to hand with depression in most of the cases. Anxiety disorder is related with increased risk of coronary heart disease, atherosclerosis, and metabolic disorders. Because these conditions have low grade of inflammatory component, the researchers can predict the modification of inflammatory pathways, and immune component may be a reason for the progression of anxiety disorders.

The drugs like SSRIs are useful in relieving anxiety via modification of inflammatory pathways. Tricyclic antidepressants like imipramine and clomipramine also found to be effective in anxiety and obsessive-compulsive disorders, respectively, via modification in the levels of pro-inflammatory markers. High levels of phobic anxiety state are associated with increased leptin and inflammatory marker levels [159]. Oxidative stress and HPA axis dysregulation are also seen in case of anxiety disorders and be involved in inflammation and immune component. The drugs target oxidative stress pathways like anti-oxidants such as vitamin C and E, resveratrol, curcumin, etc., which are natural substances and can be useful to treat CNS disorders via influence of inflammation [160]. CRF-1 receptor antagonists that target HPA axis also have found to show beneficial effects in depression and anxiety [161].

#### 18.4.1.5 Anti-Alzheimer's Drugs on Inflammation

The pathological progression of AD involves neurodegeneration following the deposition of β-amyloid (Aβ) plaques and neurofibrillary tangles in vulnerable brain regions [162]. Inflammation is also one of the important components to play a role in progression of AD. In cell line and mouse study of AD, an NSAID subset of ibuprofen, indomethacin, and sulindac has been demonstrated to reduce the production of the 42 residue β-amyloid peptide independently of changes in cyclooxygenase activity [163]. The main category of drugs used in the AD is choline-esterase inhibitors such as tacrine, rivastigmine, donepezil, etc. In addition, immunotherapies induce circulating antibodies to the Aβ peptide so that they can either bind and sequester the circulating Aβ from the blood [164], inhibit Aβ fibrillogenesis or toxic oligomer formation [165], or bind to plaques in the brain and stimulate Fc-γ receptor-mediated phagocytosis by microglia [166]. It looks that the above mechanisms could be associated with immune clearance of Aβ in mice. However, data from some other study indicate that antibody-mediated mobilization of Aβ from plaques has the potential to transform Aβ into more toxic and inflammatory soluble oligomeric forms [167]. The above findings opened the concepts of immune stimulation as therapeutic approach. However, the same approach thought to be pathogenic earlier.

Herbal drugs like resveratrol, curcumin, and innate vitamins like vitamins C and E also produced beneficial effect in patients of AD [162]. One more finding demonstrated that locking the Aβ-binding receptor for advanced glycation end products (RAGE) on Aβ- treated human microglia has significant anti-inflammatory properties. RAGE, which is upregulated on a number of cell types in AD brains including microglia, astrocytes, vascular cells, and neurons, is currently a drug target

for AD and a number of other vascular and inflammatory diseases [168]. Induction of microglia that used an expression leads to induction of various pro-inflammatory markers such as IL-1β, IL-8, MCP- 1, MMP, IL-6, Cox-2, etc. Not only changes in the expression of genes associated with human microglia are markedly pro-inflammatory but also some potential anti-inflammatory mediators are also stimulated such as IL-1 receptor antagonist, somatostatin receptor-2, vitamin D receptor, endothelial cell protein C receptor, and adenosine 2A receptor, etc. [169].

#### 18.4.1.6 Antipsychotic Drugs on Inflammation

The effect of antipsychotic drugs on inflammation is controversial. Previous studies have demonstrated that polymorphism in genes is associated with modification of immune system in schizophrenia. Disruption of cytokine networks and change and increase in circulating peripheral immune cells are also observed in psychoses [170, 171].

The in vitro studies on antipsychotics are also have shown less clear-cut idea, showing pro- and anti-inflammatory activity for the same antipsychotic agent (haloperidol, clozapine, and risperidone) across different studies. Al-Amin et al., 2013, have demonstrated that haloperidol and quetiapine significantly increased the IL-4 levels ($p < 0.05$) in LPS-stimulated PBMC cultures, while clozapine and quetiapine predominantly enhanced the IL-4 levels ($p < 0.05$) in poly(I:C)-stimulated PBMC cultures. Only treatment with haloperidol resulted in a significant increase in IL-10 production ($p < 0.05$) in LPS-stimulated PBMC cultures, whereas clozapine, quetiapine, and risperidone treatment markedly increased IL-10 production ($p < 0.05$) in poly(I:C)-stimulated PBMC cultures [172]. Immunomodulation approaches include the use of NSAIDs, antioxidants, antioxidant vitamins, some herbal products, and other neuroprotection agents that inhibit pro-inflammatory processes. Clozapine found to blunt inflammatory responses and certain biological agents to antagonize specific immune mediators such as the cytokines. Combination of synthetic molecule of biological agent may be a useful approach for the treatment of schizophrenia via modification of inflammation and immune function. In addition, patients with high levels of C-reactive protein, IL-6, IFN-γ, TNF-α, and genetic polymorphisms of cytokines with schizophrenia can be targeted for personalized medicine [173]. The personalize medicine should target the inflammatory pathways for the treatment of schizophrenia in such patients. In addition, clozapine reduced the level of poly(I:C)-activated NLRP3 expression by 57%. The result gives an idea that clozapine might produce its anti-inflammatory properties via inhibiting NLRP3 inflammasome [174].

## 18.5 Conclusion

Inflammation and immune system are highly integrated and involved in the pathogenesis of neuropsychiatric disorders. We can target inflammation and immune components for the treatment of the disorders like depression, anxiety and stress disorders, AD, and schizophrenia. Researchers observed that levels of inflammatory

markers in brain and blood are increased in the pathogenesis of abovementioned disorders. Hence, targeting the inflammatory pathway and immune component may be an attractive strategy. Some of the standard drugs used in the treatment of psychiatric disorders also target these pathways indirectly or directly. For example, some anti-oxidant vitamins or drugs like curcumin and resveratrol targets the oxidative stress pathways, which also results in pronounced inflammation. However, some more studies are required to reach in any fruitful conclusion.

## References


1. Ghaemi SN. A new nomenclature for psychotropic drugs. J Clin Psychopharmacol. 2015;35 (4):428–33.
2. Valenzuela CF, Puglia MP, Zucca S. Focus on: neurotransmitter systems. Alcohol Res Health. 2011;34(1):106–20.
3. Haroon E, Raison CL, Miller AH. Psychoneuroimmunology meets neuropsychopharmacology: translational implications of the impact of inflammation on behavior. Neuropsychopharmacology. 2012;37(1):137–62.
4. Miller AH, Haroon E, Raison CL, Felger JC. Cytokine targets in the brain: impact on neurotransmitters and neurocircuits. Depress Anxiety. 2013;30(4):297–306.
5. Westfall S, Pasinetti GM. The gut microbiota links dietary polyphenols with management of psychiatric mood disorders. Front Neurosci. 2019;13:1196.
6. Bennett FC, Molofsky AV. The immune system and psychiatric disease: a basic science perspective. Clin Exp Immunol. 2019;197(3):294–307.
7. WHO Website. https://www.who.int/news-room/fact-sheets/detail/depression. Accessed 10 Oct 2021.
8. Boku S, Nakagawa S, Toda H, Hishimoto A. Neural basis of major depressive disorder: beyond monoamine hypothesis. Psychiatry Clin Neurosci. 2018;72:3–12.
9. Heninger GR, Delgado PL, Charney DS. The revised monoamine theory of depression: a modulatory role for monoamines, based on new findings from monoamine depletion experiments in humans. Pharmacopsychiatry. 1996;29:2–11.
10. Lener MS, Niciu MJ, Ballard ED, Park M, Park LT, Nugent AC, Zarate CA Jr. Glutamate and gamma-aminobutyric acid Systems in the Pathophysiology of major depression and antidepressant response to ketamine. Biol Psychiatry. 2017;81(10):886–97.
11. Liu W, Ge T, Leng Y, Pan Z, Fan J, Yang W, Cui R. The role of neural plasticity in depression: from hippocampus to prefrontal cortex. Neural Plast. 2017;2017:6871089.
12. Shadrina M, Bondarenko EA, Slominsky PA. Genetics factors in major depression disease. Front Psych. 2018;9:334.
13. Michel TM, Pülschen D, Thome J. The role of oxidative stress in depressive disorders. Curr Pharm Des. 2012;18:5890–9.
14. Raison CL, Miller AH. Is depression an inflammatory disorder? Curr Psychiatry Rep. 2011;13 (6):467–75.
15. https://www.medicalnewstoday.com/articles/anxiety-and-inflammation-is-there-a-link. Accessed 15 Oct 2021.
16. Kessler RC, Berglund P, Demler O, Jin R, Koretz D, Merikangas KR, Rush AJ, Walters EE, Wang PS. National Comorbidity Survey Replication. The epidemiology of major depressive disorder: results from the National Comorbidity Survey Replication (NCS-R). JAMA. 2003;289(23):3095–105.
17. Carter RM, Wittchen HU, Pfister H, Kessler RC. One-year prevalence of subthreshold and threshold DSM-IV generalized anxiety disorder in a nationally representative sample. Depress Anxiety. 2001;13:78–88.

18. Ressler KJ, Nemeroff CB. Role of serotonergic and noradrenergic systems in the pathophysiology of depression and anxiety disorders. Depress Anxiety. 2000;12(Suppl 1):2–19.
19. Nuss P. Anxiety disorders and GABA neurotransmission: a disturbance of modulation. Neuropsychiatr Dis Treat. 2015;11:165–75.
20. Bhatt S, Devadoss T, Manjula SN, Rajangam J. 5-$HT_3$ receptor antagonism a potential therapeutic approach for the treatment of depression and other disorders. Curr Neuropharmacol. 2021;19(9):1545–59.
21. Danese A, Lewis J. Psychoneuroimmunology of early-life stress: the hidden wounds of childhood trauma? Neuropsychopharmacology. 2017;42:99–114.
22. Salim S, Chugh G, Asghar M. Inflammation in anxiety. Adv Protein Chem Struct Biol. 2012;88:1–25.
23. Bandelow B, Michaelis S, Wedekind D. Treatment of anxiety disorders. Dialogues Clin Neurosci. 2017;19(2):93–107.
24. Locke AB, Kirst N, Shultz CG. Diagnosis and management of generalized anxiety disorder and panic disorder in adults. Am Fam Physician. 2015;91(9):617–24.
25. Jakubovski E, Johnson JA, Nasir M, Müller-Vahl K, Bloch MH. Systematic review and meta-analysis: dose-response curve of SSRIs and SNRIs in anxiety disorders. Depress Anxiety. 2019;36(3):198–212.
26. Alzheimer A. Ueber einen eigenartigen schweren Erkrankungsprozess der Hirnrinde. Neurol Central. 1906;25:1134.
27. Probst A, Langui D, Ulrich J. Alzheimer's disease: a description of the structural lesions. Brain Pathol. 1991;1(4):229–39.
28. Ginhoux F, Greter M, Leboeuf M, et al. Fate mapping analysis reveals that adult microglia derive from primitive macrophages. Science. 2010;330:841–5.
29. Braak H, Braak E. Neuropathological stageing of Alzheimer-related changes. Acta Neuropathol. 1991;82:239–59.
30. Heneka MT, Kummer MP, Latz E. Innate immune activation in neurodegenerative disease. Nat Rev Immunol. 2014;14:463–77.
31. Shariati SA, De SB. Redundancy and divergence in the amyloid precursor protein family. FEBS Lett. 2013;587(13):2036–45.
32. Delgado A, Cholevas C, Theoharides TC. Neuroinflammation in Alzheimer's disease and beneficial action of luteolin. Biofactors. 2021;47(2):207–17.
33. Wilkins HM, Swerdlow RH. Amyloid precursor protein processing and bioenergetics. Brain Res Bull. 2017;133:71–9.
34. Karran E, De SB. The amyloid cascade hypothesis: are we poised for success or failure? J Neurochem. 2016;139(Suppl 2):237–52.
35. Rocha-Souto B, Scotton TC, Coma M, et al. Brain oligomeric beta-amyloid but not total amyloid plaque burden correlates with neuronal loss and astrocyte inflammatory response in amyloid precursor protein/tau transgenic mice. J Neuropathol Exp Neurol. 2011;70(5):360–76.
36. Gong CX, Iqbal K. Hyperphosphorylation of microtubule associated protein tau: a promising therapeutic target for Alzheimer disease. Curr Med Chem. 2008;15(23):2321–8.
37. Ebneth A, Godemann R, Stamer K, Illenberger S, Trinczek B, Mandelkow E. Overexpression of tau protein inhibits kinesindependent trafficking of vesicles, mitochondria, and endoplasmic reticulum: implications for Alzheimer's disease. J Cell Biol. 1998;143(3):777–94.
38. Guo T, Noble W, Hanger DP. Roles of tau protein in health and disease. Acta Neuropathol. 2017;133(5):665–704.
39. Alonso AD, Grundke-Iqbal I, Barra HS, Iqbal K. Abnormal phosphorylation of tau and the mechanism of Alzheimer neurofibrillary degeneration: sequestration of microtubule-associated proteins 1 and 2 and the disassembly of microtubules by the abnormal tau. Proc Natl Acad Sci U S A. 1997;94:298–303.
40. Iqbal K, Liu F, Gong C-X, Grundke-Iqbal I. Tau in Alzheimer disease and related tauopathies. Curr Alzheimer Res. 2010;7:656–64.

41. Köpke E, Tung YC, Shaikh S, Alonso AC, Iqbal K, Grundke-Iqbal I. Microtubule-associated protein tau. Abnormal phosphorylation of a non-paired helical filament pool in Alzheimer disease. J Biol Chem. 1993;268:24374–84.
42. Lippens G, Sillen A, Landrieu I, Amniai L, Sibille N, Barbier P, et al. Tau aggregation in Alzheimer's disease. Prion. 2007;1:21–5.
43. Simi G, Babi Leko M, Wray S, Harrington C, Delalle I, Jovanov-Milošević N, et al. Tau protein hyperphosphorylation and aggregation in Alzheimer's disease and other tauopathies, and possible neuroprotective strategies. Biomol Ther. 2016;6, 6
44. Hardy JA, Higgins GA. Alzheimer's disease: the amyloid cascade hypothesis. Science. 1992;256(5054):184–5.
45. Dickson DW. The pathogenesis of senile plaques. J Neuropathol Exp Neurol. 1997;56(4): 321–39.
46. Selkoe DJ. Translating cell biology into therapeutic advances in Alzheimer's disease. Nature. 1999;399(6738 Suppl):A23–31.
47. Lue LF, Kuo YM, Roher AE, Brachova L, Shen Y, Sue L, et al. Soluble amyloid beta peptide concentration as a predictor of synaptic change in Alzheimer's disease. Am J Pathol. 1999;155 (3):853–62.
48. Wang J, Dickson DW, Trojanowski JQ, Lee VM. The levels of soluble versus insoluble brain Abeta distinguish Alzheimer's disease from normal and pathologic aging. Exp Neurol. 1999;158(2):328–37.
49. Honig LS, Vellas B, Woodward M, Boada M, Bullock R, Borrie M, et al. Trial of solanezumab for mild dementia due to Alzheimer's disease. N Engl J Med. 2018;378(4):321–30.
50. Selkoe DJ. Alzheimer disease and aducanumab: adjusting our approach. Nat Rev Neurol. 2019;15(7):365–6.
51. Morris GP, Clark IA, Vissel B. Inconsistencies and controversies surrounding the amyloid hypothesis of Alzheimer's disease. Acta Neuropathol Commun. 2014;2:135.
52. Heppner FL, Ransohoff RM, Becher B. Immune attack: the role of inflammation in Alzheimer disease. Nat Rev Neurosci. 2015;16(6):358–72.
53. Kinney JW, Bemiller SM, Murtishaw AS, Leisgang AM, Salazar AM, Lamb BT. Inflammation as a central mechanism in Alzheimer's disease. Alzheimers Dement (N Y). 2018;4:575–90.
54. Garwood CJ, Pooler AM, Atherton J, Hanger DP, Noble W. Astrocytes are important mediators of ab-induced neurotoxicity and tau phosphorylation in primary culture. Cell Death Dis. 2011;2:e167.
55. Kitazawa M, Oddo S, Yamasaki TR, Green KN, LaFerla FM. Lipopolysaccharide-induced inflammation exacerbates tau pathology by a cyclin-dependent kinase 5-mediated pathway in a transgenic model of Alzheimer's disease. J Neurosci. 2005;25:8843–53.
56. Kitazawa M, Yamasaki TR, Laferla FM. Microglia as a potential bridge between the amyloid b-peptide and tau. Ann N Y Acad Sci. 2004;1035:85–103.
57. Aricioglu F, Ozkartal CS, Unal G, Dursun SD. Neuroinflammation in schizophrenia: a critical review and the future. Bull Clin Psychopharmacol. 2016;26(4):329–444.
58. Meyer U. Anti-inflammatory signaling in schizophrenia. Brain Behav Immun. 2011;25(8): 1507–18.
59. Howes OD, Kapur S. The dopamine hypothesis of schizophrenia: version III--the final common pathway. Schizophr Bull. 2009;35:549–62.
60. Miyamoto S, Duncan GE, Marx CE, Lieberman JA. Treatments for schizophrenia: a critical review of pharmacology and mechanisms of action of antipsychotic drugs. Mol Psychiatry. 2005;10:79–104.
61. Foussias G, Remington G. Antipsychotics and schizophrenia: from efficacy and effectiveness to clinical decision- making. Can J Psychiatr. 2010;55:117–25.
62. Altamura AC, Pozzoli S, Fiorentini A, Dell'Osso B. Neurodevelopment and inflammatory patterns in schizophrenia in relation to pathophysiology. Prog Neuro-Psychopharmacol Biol Psychiatry. 2013;42(8):63–70.

63. Brown AS. Epidemiologic studies of exposure to prenatal infection and risk of schizophrenia and autism. Dev Neurobiol. 2012;72(10):1272–6.
64. Meyer U, Feldon J. To poly(I:C) or not to poly(I:C): advancing preclinical schizophrenia research through the use of prenatal immune activation models. Neuropharmacology. 2012;62 (3):1308–21.
65. Hong J, Bang M. Anti-inflammatory strategies for schizophrenia: a review of evidence for therapeutic applications and drug repurposing. Clin Psychopharmacol Neurosci. 2020;18(1): 10–24.
66. Müller N, Weidinger E, Leitner B, Schwarz MJ. The role of inflammation in schizophrenia. Front Neurosci. 2015;9:372.
67. Khandaker GM, Cousins L, Deakin J, Lennox BR, Yolken R, Jones PB. Inflammation and immunity in schizophrenia: implications for pathophysiology and treatment. Lancet Psychiatry. 2015;2(3):258–70.
68. https://americanaddictioncenters.org/trauma-stressor-related-disorders. Accessed 15 Oct 2021.
69. Bhatt S, Mahesh R, Jindal A, Devadoss T. Neuropharmacological and neurochemical evaluation of N-n-propyl-3-ethoxyquinoxaline-2-carboxamide (6n): a novel serotonergic 5-HT3 receptor antagonist for co-morbid antidepressant- and anxiolytic-like potential using traumatic brain injury model in rats. J Basic Clin Physiol Pharmacol. 2017;28(2):93–100.
70. Ng SY, Lee AYW. Traumatic brain injuries: pathophysiology and potential therapeutic targets. Front Cell Neurosci. 2019;13:528.
71. Beurel E, Toups M, Nemeroff CB. The bidirectional relationship of depression and inflammation: double trouble. Neuron. 2020;107(2):234–56.
72. Kim SU, de Vellis J. Microglia in health and disease. J Neurosci Res. 2005;81(3):302–13.
73. Yirmiya R, Rimmerman N, Reshef R. Depression as a microglial disease. Trends Neurosci. 2015;38(10):637–58.
74. Bakunina N, Pariante CM, Zunszain PA. Immune mechanisms linked to depression via oxidative stress and neuroprogression. Immunology. 2015;144(3):365–73.
75. Lima Giacobbo B, Doorduin J, Klein HC, Dierckx RAJO, Bromberg E, de Vries EFJ. Brain-derived neurotrophic factor in brain disorders: focus on Neuroinflammation. Mol Neurobiol. 2019;56(5):3295–312.
76. Wooff Y, Man SM, Aggio-Bruce R, Natoli R, Fernando N. IL-1 family members mediate cell death, inflammation and angiogenesis in retinal degenerative diseases. Front Immunol. 2019;10:1618.
77. Kopschina Feltes P, Doorduin J, Klein HC, Juárez-Orozco LE, Dierckx RA, Moriguchi-Jeckel CM, de Vries EF. Anti-inflammatory treatment for major depressive disorder: implications for patients with an elevated immune profile and non-responders to standard antidepressant therapy. J Psychopharmacol. 2017;31(9):1149–65.
78. Raison CL, Capuron L, Miller AH. Cytokines sing the blues: inflammation and the pathogenesis of depression. Trends Immunol. 2006;27:24–31.
79. Howren MB, Lamkin DM, Suls J. Associations of depression with C-reactive protein, IL-1, and IL-6: a meta-analysis. Psychosom Med. 2009;71:171–86.
80. Dowlati Y, Herrmann N, Swardfager W, Liu H, Sham L, Reim EK, Lanctôt KL. A meta-analysis of cytokines in major depression. Biol Psychiatry. 2010;67(5):446–57.
81. Bhatt S, Nagappa AN, Patil CR. Role of oxidative stress in depression. Drug Discov Today. 2020;25(7):1270–6.
82. Valkanova V, Ebmeier KP, Allan CL. CRP, IL-6 and depression: a systematic review and meta-analysis of longitudinal studies. J Affect Disord. 2013;150:736–44.
83. Allen AP, Kennedy PJ, Cryan JF, Dinan TG, Clarke G. Biological and psychological markers of stress in humans: focus on the Trier social stress test. Neurosci Biobehav Rev. 2014;38:94–124.
84. Slavich GM, Irwin MR. From stress to inflammation and major depressive disorder: a social signal transduction theory of depression. Psychol Bull. 2014;140:774.

85. Rohleder N. Stimulation of systemic low-grade inflammation by psychosocial stress. Psychosom Med. 2014;76:181–9.
86. Pace TW, Mletzko TC, Alagbe O, Musselman DL, Nemeroff CB, Miller AH, Heim CM. Increased stress-induced inflammatory responses in male patients with major depression and increased early life stress. Am J Psychiatry. 2006;163(9):1630–3.
87. Calcia MA, Bonsall DR, Bloomfield PS, Selvaraj S, Barichello T, Howes OD. Stress and neuroinflammation: a systematic review of the effects of stress on microglia and the implications for mental illness. Psychopharmacology. 2016;233(9):1637–50.
88. Quan N, Banks WA. Brain-immune communication pathways. Brain Behav Immun. 2007;21:727–35.
89. Sorrells SF, Caso JR, Munhoz CD, Sapolsky RM. The stressed CNS: when glucocorticoids aggravate inflammation. Neuron. 2009;64(1):33–9.
90. Liu YZ, Wang YX, Jiang CL. Inflammation: the common pathway of stress-related diseases. Front Hum Neurosci. 2017;11:316.
91. Hou R, Garner M, Holmes C, Osmond C, Teeling J, Lau L, Baldwin DS. Peripheral inflammatory cytokines and immune balance in generalised anxiety disorder: case-controlled study. Brain Behav Immun. 2017;62:212–8.
92. Morales I, Guzmán-Martínez L, Cerda-Troncoso C, Farías GA, Maccioni RB. Neuroinflammation in the pathogenesis of Alzheimer's disease. A rational framework for the search of novel therapeutic approaches. Front Cell Neurosci. 2014;8:112.
93. McNaull BB, Todd S, McGuinness B, Passmore AP. Inflammation and anti-inflammatory strategies for Alzheimer's disease – a mini-review. Gerontology. 2010;56:3–14.
94. Bagyinszky E, Van Giau V, Shim K, Suk K, An SSA, Kim S. Role of inflammatory molecules in the Alzheimer's disease progression and diagnosis. J Neurol Sci. 2017;376:242–54.
95. DiSabato DJ, Quan N, Godbout JP. Neuroinflammation: the devil is in the details. J Neurochem. 2016;139(Suppl 2):136–53.
96. Streit WJ, Mrak RE, Griffin WS. Microglia and neuroinflammation: a pathological perspective. J Neuroinflammation. 2004;1(1):14.
97. Shastri A, Bonifati DM, Kishore U. Innate immunity and neuroinflammation. Mediators Inflamm. 2013;2013:342931.
98. Ahmad MH, Fatima M, Mondal AC. Influence of microglia and astrocyte activation in the neuroinflammatory pathogenesis of Alzheimer's disease: rational insights for the therapeutic approaches. J Clin Neurosci. 2019;59:6–11.
99. Agostinho P, Cunha RA, Oliveira C. Neuroinflammation, oxidative stress and the pathogenesis of Alzheimer's disease. Curr Pharm Des. 2010;16:2766–78.
100. Avila-Muñoz E, Arias C. When astrocytes become harmful: functional and inflammatory responses that contribute to Alzheimer's disease. Ageing Res Rev. 2014;18:29–40.
101. Chen CH, Zhou W, Liu S, Deng Y, Cai F, Tone M, et al. Increased NF-kappa B signalling up-regulates BACE1 expression and its therapeutic potential in Alzheimer's disease. Int J Neuropsychopharmacol. 2012;15(1):77–90.
102. Webers A, Heneka MT, Gleeson PA. The role of innate immune responses and neuroinflammation in amyloid accumulation and progression of Alzheimer's disease. Immunol Cell Biol. 2020;98(1):28–41.
103. Lee JW, Lee YK, Yuk DY, et al. Neuro-inflammation induced by lipopolysaccharide causes cognitive impairment through enhancement of beta-amyloid generation. J Neuroinflammation. 2008;5:37.
104. Heneka MT. Inflammasome activation and innate immunity in Alzheimer's disease. Brain Pathol. 2017;27:220–2.
105. Ye L, Huang Y, Zhao L, et al. IL-1β and TNF-α induce neurotoxicity through glutamate production: a potential role for neuronal glutaminase. J Neurochem. 2013;125:897–908.
106. Fillit H, Ding WH, Buee L, Kalman J, Altstiel L, Lawlor B, et al. Elevated circulating tumor necrosis factor levels in Alzheimer's disease. Neurosci Lett. 1991;129(2):318–20.

107. Strauss S, Bauer J, Ganter U, Jonas U, Berger M, Volk B. Detection of interleukin-6 and alpha 2-macroglobulin immunoreactivity in cortex and hippocampus of Alzheimer's disease patients. Lab Investig. 1992;66(2):223–30.
108. Griffin WS, Stanley LC, Ling C, White L, MacLeod V, Perrot LJ, et al. Brain interleukin 1 and S-100 immunoreactivity are elevated in down syndrome and Alzheimer disease. Proc Natl Acad Sci U S A. 1989;86(19):7611–5.
109. Grammas P, Ovase R. Inflammatory factors are elevated in brain microvessels in Alzheimer's disease. Neurobiol Aging. 2001;22(6):837–42.
110. Rogers J, Luber-Narod J, Styren SD, Civin WH. Expression of immune system-associated antigens by cells of the human central nervous system: relationship to the pathology of Alzheimer's disease. Neurobiol Aging. 1988;9(4):339–49.
111. Miller BJ, Buckley P, Seabolt W, Mellor A, Kirkpatrick B. Meta-analysis of cytokine alterations in schizophrenia: clinical status and antipsychotic effects. Biol Psychiatry. 2011;70:663–71.
112. Bolu A, Aydın MS, Akgün A, Coşkun A, Garip B, Öznur T, et al. Serum levels of high sensitivity c-reactive protein in drug-naïve first-episode psychosis and acute exacerbation of schizophrenia. Clin Psychopharmacol Neurosci. 2019;17:244–9.
113. Monji A, Kato TA, Mizoguchi Y, Horikawa H, Seki Y, Kasai M, et al. Neuroinflammation in schizophrenia especially focused on the role of microglia. Prog Neuro-Psychopharmacol Biol Psychiatry. 2013;42:115–21.
114. Furukawa H, Del Rey A, Monge-Arditi G, Besedovsky HO. Interleukin-1, but not stress, stimulates glucocorticoid output during early postnatal life in mice. Ann N Y Acad Sci. 1998;840:117–22.
115. Nair A, Bonneau RH. Stress-induced elevation of glucocorticoids increases microglia proliferation through NMDA receptor activation. J Neuroimmunol. 2006;171:72–85.
116. Sparkman NL, Johnson RW. Neuroinflammation associated with aging sensitizes the brain to the effects of infection or stress. Neuroimmunomodulation. 2008;15:323–30.
117. Frank MG, Baratta MV, Sprunger DB, Watkins LR, Maier SF. Microglia serve as a neuroimmune substrate for stress-inducedpotentiation of CNS pro-inflammatory cytokine responses. Brain Behav Immun. 2007;21:47–59.
118. Zhou D, Kusnecov AW, Shurin MR, DePaoli M, Rabin BS. Exposure to physical and psychological stressors elevates plasma interleukin 6: relationship to the activation of hypothalamic pituitary-adrenal axis. Endocrinology. 1993;133:2523–30.
119. Howes OD, McCutcheon R. Inflammation and the neural diathesis-stress hypothesis of schizophrenia: a reconceptualization. Transl Psychiatry. 2017;7:e1024.
120. Zalcman S, Murray L, Dyck DG, Greenberg AH, Nance DM. Interleukin-2 and -6 induce behavioral-activating effects in mice. Brain Res. 1998;811:111–21.
121. Zalcman S, Savina I, Wise RA. Interleukin-6 increases sensitivity to the locomotor-stimulating effects of amphetamine in rats. Brain Res. 1999;847:276–83.
122. Behrens MM, Ali SS, Dugan LL. Interleukin-6 mediates the increase in NADPH-oxidase in the ketamine model of schizophrenia. J Neurosci. 2008;28:13957–66.
123. Block ML, Hong JS. Microglia and inflammation-mediated neurodegeneration: multiple triggers with a common mechanism. Prog Neurobiol. 2005;76:77–98.
124. Parrott JM, O'Connor JC. Kynurenine 3-monooxygenase: an influential mediator of neuropathology. Front Psych. 2015;6:116.
125. Stone TW, Darlington LG. Endogenous kynurenines as targets for drug discovery and development. Nat Rev Drug Discov. 2002;1:609–20.
126. Iaccarino HF, Suckow RF, Xie S, Bucci DJ. The effect of transient increases in kynurenic acid and quinolinic acid levels early in life on behavior in adulthood: implications for schizophrenia. Schizophr Res. 2013;150:392–7.
127. Larsson MK, Faka A, Bhat M, Imbeault S, Goiny M, Orhan F, et al. Repeated LPS injection induces distinct changes in the kynurenine pathway in mice. Neurochem Res. 2016;41:2243–55.

128. Pérez-Neri I, Ramírez-Bermúdez J, Montes S, Ríos C. Possible mechanisms of neurodegeneration in schizophrenia. Neurochem Res. 2006;31:1279–94.
129. Trépanier MO, Hopperton KE, Mizrahi R, Mechawar N, Bazinet RP. Postmortem evidence of cerebral inflammation in schizophrenia: a systematic review. Mol Psychiatry. 2016;21:1009–26.
130. Van Kesteren CF, Gremmels H, de Witte LD, Hol EM, Van Gool AR, Falkai PG, et al. Immune involvement in the pathogenesis of schizophrenia: a meta-analysis on postmortem brain studies. Transl Psychiatry. 2017;7:e1075.
131. Doorduin J, de Vries EF, Willemsen AT, de Groot JC, Dierckx RA, Klein HC. Neuroinflammation in schizophrenia-related psychosis: a PET study. J Nucl Med. 2009;50:1801–7.
132. Van Berckel BN, Bossong MG, Boellaard R, Kloet R, Schuitemaker A, Caspers E, et al. Microglia activation in recent- onset schizophrenia: a quantitative (R)-[11C]PK1195positron emission tomography study. Biol Psychiatry. 2008;64:820–2.
133. Bloomfield PS, Selvaraj S, Veronese M, Rizzo G, Bertoldo A, Owen DR, et al. Microglial activity in people at ultra-highrisk of psychosis and in schizophrenia: an [(11)C]PBR28 PET brain imaging study. Am J Psychiatry. 2016;173:44–52.
134. Kim YK, Myint AM, Lee BH, Han CS, Lee HJ, Kim DJ, et al. Th1, Th2 and Th3 cytokine alteration in schizophrenia. Prog Neuro-Psychopharmacol Biol Psychiatry. 2004;28(7): 1129–34.
135. Martínez-Gras I, García-Sánchez F, Guaza C, Rodríguez-Jiménez R, Andrés-Esteban E, Palomo T, et al. Altered immune function in unaffected first-degree biological relatives of schizophrenia patients. Psychiatry Res. 2012;200(2–3):1022–5.
136. Meyer U, Schwarz MJ, Müller N. Inflammatory processes in schizophrenia: a promising neuroimmunological target for the treatment of negative/cognitive symptoms and beyond. Pharmacol Ther. 2011;132(1):96–110.
137. Müller N, Wagner JK, Krause D, Weidinger E, Wildenauer A, Obermeier M, et al. Impaired monocyte activation inschizophrenia. Psychiatry Res. 2012;198(3):341–6.
138. Rothermundt M, Ahn JN, Jörgens S. S100B in schizophrenia: an update. Gen Physiol Biophys. 2009;28:76–81.
139. De Witte L, Tomasik J, Schwarz E, Guest PC, Rahmoune H, Kahn RS, et al. Cytokine alterations in first-episode schizophrenia patients before and after antipsychotic treatment. Schizophr Res. 2014;154(1–3):23–9.
140. Maes M, Meltzer HY, Bosmans E. Immune-inflammatory markers in schizophrenia: comparison to normal controls and effects of clozapine. Acta Psychiatr Scand. 1994;89(5):346–51.
141. Müller N, Bechter K. The mild encephalitis concept for psychiatric disorders revisited in the light of current psychoneuroimmunological findings. Neurol Psychiatry Brain Res. 2013;19 (3):87–101.
142. Dantzer R. Cytokine-induced sickness behaviour: a neuroimmune response to activation of innate immunity. Eur J Pharmacol. 2004;1(500):399–411.
143. Layé S, Gheusi G, Cremona S, Combe C, Kelley K, Dantzer R, et al. Endogenous brain IL-1 mediates LPS-induced anorexia and hypothalamic cytokine expression. Am J Physiol Regul Integr Comp Physiol. 2000;279(1):93–8.
144. Lotocki G, de Rivero Vaccari JP, Perez ER, Sanchez-Molano J, Furones-Alonso O, Bramlett HM, Dietrich WD. Alterations in blood-brain barrier permeability to large and small molecules and leukocyte accumulation after traumatic brain injury: effects of post-traumatic hypothermia. J Neurotrauma. 2009;26(7):1123–34.
145. Goodman JC, Van M, Gopinath SP, Robertson CS. Pro-inflammatory and pro-apoptotic elements of the neuroinflammatory response are activated in traumatic brain injury. Acta Neurochir Suppl. 2009;102:437–9.
146. Frugier T, Morganti-Kossmann MC, O'Reilly D, Mclean CA. In situ detection of inflammatory mediators in post mortem human brain tissue after traumatic injury. J Neurotrauma. 2009;27:497–507.

147. Semple BD, Bye N, Rancan M, Ziebell JM, Morganti-Kossmann MC. Role of CCL2 (MCP-1) in traumatic brain injury (TBI): evidence from severe TBI patients and CCL2−/− mice. J Cereb Blood Flow Metab. 2010;30:769–82.
148. Buttram SD, Wisniewski SR, Jackson EK, Adelson PD, Feldman K, Bayir H, Berger RP, Clark RS, Kochanek PM. Multiplex assessment of cytokine and chemokine levels in cerebrospinal fluid following severe pediatric traumatic brain injury: effects of moderate hypothermia. J Neurotrauma. 2007;24(11):1707–17.
149. Rancan M, Otto VI, Hans VH, Gerlach I, Jork R, Trentz O, et al. Upregulation of ICAM-1 and MCP-1 but not of MIP-2 and sensorimotor deficit in response to traumatic axonal injury in rats. J Neurosci Res. 2001;63:438–46.
150. Johnson VE, Stewart JE, Begbie FD, Trojanowski JQ, Smith DH, Stewart W. Inflammation and white matter degeneration persist for years after a single traumatic brain injury. Brain. 2013;136:28–42.
151. Baumeister D, Ciufolini S, Mondelli V. Effects of psychotropic drugs on inflammation: consequence or mediator of therapeutic effects in psychiatric treatment? Psychopharmacology. 2016;233(9):1575–89.
152. Hashioka S, McGeer PL, Monji A, Kanba S. Anti-inflammatory effects of antidepressants: possibilities for preventives against Alzheimer's disease. Cent Nerv Syst Agents Med Chem. 2009;9(1):12–9.
153. Hannestad J, DellaGioia N, Bloch M. The effect of antidepressant medication treatment on serum levels of inflammatory cytokines: a meta-analysis. Neuropsychopharmacology. 2011;36:2452–9.
154. Arteaga-Henríquez G, Simon MS, Burger B, Weidinger E, Wijkhuijs A, Arolt V, Birkenhager TK, Musil R, Müller N, Drexhage HA. Low-grade inflammation as a predictor of antidepressant and anti-inflammatory therapy response in MDD patients: a systematic review of the literature in combination with an analysis of experimental data collected in the EU-MOODINFLAME consortium. Front Psychiatry. 2019;10:458.
155. Ohgi Y, Futamura T, Kikuchi T, Hashimoto K. Effects of antidepressants on alternations in serum cytokines and depressive-like behavior in mice after lipopolysaccharide administration. Pharmacol Biochem Behav. 2013;103(4):853–9.
156. Bhatt S, Mahesh R, Devadoss T, Jindal A. Neuropharmacological evaluation of a novel 5-HT3 receptor antagonist (4-benzylpiperazin-1-yl)(3-methoxyquinoxalin-2-yl) methanone (6g) on lipopolysaccharide-induced anxiety models in mice. J Basic Clin Physiol Pharmacol. 2017;28(2):101–6.
157. Sacre S, Jaxa-Chamiec A, Low CMR, Chamberlain G, Tralau-Stewart C. Structural modification of the antidepressant Mianserin suggests that its anti-inflammatory activity may be independent of 5-Hydroxytryptamine receptors. Front Immunol. 2019;10:1167.
158. Groen RN, Ryan O, Wigman JTW, Riese H, Penninx BWJH, Giltay EJ, Wichers M, Hartman CA. Comorbidity between depression and anxiety: assessing the role of bridge mental states in dynamic psychological networks. BMC Med. 2020;18(1):308.
159. Murrough JW, Yaqubi S, Sayed S, Charney DS. Emerging drugs for the treatment of anxiety. Expert Opin Emerg Drugs. 2015;20(3):393–406.
160. Lobo V, Patil A, Phatak A, Chandra N. Free radicals, antioxidants and functional foods: impact on human health. Pharmacogn Rev. 2010;4(8):118–26.
161. Taché Y, Martinez V, Wang L, Million M. CRF1 receptor signaling pathways are involved in stress-related alterations of colonic function and viscerosensitivity: implications for irritable bowel syndrome. Br J Pharmacol. 2004;141(8):1321–30.
162. Bhatt S, Puli L, Patil CR. Role of reactive oxygen species in the progression of Alzheimer's disease. Drug Discov Today. 2021;26(3):794–803.
163. Weiner HL, Selkoe DJ. Inflammation and therapeutic vaccination in CNS diseases. Nature. 2002;420:879–84.

164. DeMattos RB, Bales KR, Cummins DJ, Dodart JC, Paul SM, Holtzman DM. Peripheral anti-a beta antibody alters CNS and plasma a beta clearance and decreases brain a beta burden in a mouse model of Alzheimer's disease. Proc Natl Acad Sci U S A. 2001;98:8850–5.
165. Frenkel D, Katz O, Solomon B. Immunization against Alzheimer's beta -amyloid plaques via EFRH phage administration. Proc Natl Acad Sci U S A. 2000;97:11455–9.
166. Bard F, Cannon C, Barbour R, Burke RL, Games D, Grajeda H, Guido T, Hu K, Huang J, Johnson-Wood K, Khan K, Kholodenko D, Lee M, Lieberburg I, Motter R, Nguyen M, Soriano F, Vasquez N, Weiss K, Welch B, Seubert P, Schenk D, Yednock T. Peripherally administered antibodies against amyloid beta-peptide enter the central nervous system and reduce pathology in a mouse model of Alzheimer disease. Nat Med. 2000;6:916–9.
167. Patton RL, Kalback WM, Esh CL, Kokjohn TA, Van Vickle GD, Luehrs DC, Kuo YM, Lopez J, Brune D, Ferrer I, Masliah E, Newel AJ, Beach TG, Castano EM, Roher AE. Amyloid-beta peptide remnants in AN-1792-immunized Alzheimer's disease patients a biochemical analysis. Am J Pathol. 2006;169:1048–63.
168. Hudson BI, Schmidt AM. RAGE a novel target for drug intervention in diabetic vascular disease. Pharm Res. 2004;21:1079–86.
169. Walker D, Lue LF. Anti-inflammatory and immune therapy for Alzheimer's disease: current status and future directions. Curr Neuropharmacol. 2007;5(4):232–43.
170. Drzyzga L, Obuchowicz E, Marcinowska A, Herman ZS. Cytokines in schizophrenia and the effects of antipsychotic drugs. Brain Behav Immun. 2006;20:532–45.
171. Karanikas EP. Psycho-immunological mechanisms in schizophrenia. Psychiatrike. 2011;22: 43–52.
172. Al-Amin MM, Nasir Uddin MM, Mahmud Reza H. Effects of antipsychotics on the inflammatory response system of patients with schizophrenia in peripheral blood mononuclear cell cultures. Clin Psychopharmacol Neurosci. 2013;11(3):144–51.
173. Pandurangi AK, Buckley PF. Inflammation, antipsychotic drugs, and evidence for effectiveness of anti-inflammatory agents in schizophrenia. Curr Top Behav Neurosci. 2020;44:227–44.
174. Giridharan VV, Scaini G, Colpo GD, Doifode T, Pinjari OF, Teixeira AL, Petronilho F, Macêdo D, Quevedo J, Barichello T. Clozapine prevents poly (I:C) induced inflammation by modulating NLRP3 pathway in microglial cells. Cells. 2020;9(3):577.

# Anti-Inflammatory Effect of Traditional Chinese Medicine on the Concept of Mind-Body Interface

19

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**Abstract**

In this chapter, we conducted a systemic literature review for the anti-inflammatory effects of Traditional Chinese Medicine (TCM) applying molecular mechanisms focusing on the neuroinflammation and gut-brain axis in three neuropsychiatric disorders: major depressive disorder, Alzheimer's disease, and Parkinson's disease. We demonstrated the anti-inflammation or immunomodulation effects of TCM, including acupuncture, from basic and clinical research, including cellular and molecular approaches. In conclusion, inflammation plays a critical role in the neuropsychopathological process. At the same time, anti-inflammation seems to be the common biological pathway for the effects of TCM and acupuncture in depression, Alzheimer's disease, and Parkinson's disease.

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**Keywords**

TCM · Acupuncture · Inflammation · Immune · Depression · Alzheimer · Parkinson

## 19.1 Introduction

Neuroinflammation has an important role in neuropsychiatric diseases, including depression, Alzheimer's disease (AD), and Parkinson's disease (PD). The current evidence supports the close relationship between mind and body. Around 3000 years ago, Traditional Chinese Medicine (TCM) originated on the idea that the mind and body are interconnected [1]. In this chapter, we discussed the systemic literature review of the anti-inflammatory effects of TCM in depression, AD, and PD.

"Qing Re Yao (清熱藥)" is translated from Chinese words; it means "medicine that removes heat." In TCM theory, heat symptoms (or commonly known as "on fire (上火)") [1] indicated a similar modern concept of inflammation. Professor Cheng's team investigated 226 herbs of TCM for anti-inflammatory agents. Among the 226 herbs, 54 of them are classified as "Qing Re Yao (清熱藥)." They did the chemical analysis of six anti-inflammatory pathways—COX2, iNOS, IL-6, IFN-γ, TNF-α, and glucocorticoid—and found that 96% of "Qing Re Yao (清熱藥)" had involved at least one anti-inflammatory process. Then, they investigated the effect of combination therapy and showed the synergistic effect of multiple targets enhanced TCM efficacy.

Acupuncture was first applied around 3000 years ago and is considered one of the ancient forms of TCM, representing an ancient physiological system that believes health to be the result of harmony among bodily functions and between body and nature [2]. It showed therapeutic effects in many diseases, especially pain management. Compared to conventional treatment, acupuncture is characterized by offering a more personalized approach and better tolerance [3].

## 19.2 Methods

Two independent investigators searched the MEDLINE, CENTRAL, and EMBASE databases for eligible publications from January 1, 1980, till June 20, 2021, written in English and Chinese, using the following keywords: TCM, acupuncture, depression, major depressive disorder, Alzheimer, Parkinson, inflammation, and immune. We also checked the reference lists of relevant studies to identify any missing publications.

## 19.3 Results

### 19.3.1 Depression

We used Preferred Reporting Items for Systematic reviews and Meta-Analysis (PRISMA) guidelines for searching and listed our flowchart (Fig. 19.1). A total of 370 records were identified. Then, we excluded duplicated records, articles that didn't mention depression or major depressive disorder, animal studies, review articles, trial protocols, and articles not using TCM. We included the studies that contain inflammatory markers in depressive patients. We made a list of all the clinical research of TCM and depression for systemic review (Table 19.1).

### 19.3.2 Alzheimer's Disease

We used Preferred Reporting Items for Systematic reviews and Meta-Analysis (PRISMA) guidelines for searching and listed our flowchart (Fig. 19.2). A total of 251 records were identified. Then, we excluded duplicated records, articles that didn't mention AD, animal studies, review articles, and chemical analyses of individual TCM formulas. We included the studies that contain inflammatory markers in AD disease patients (Table 19.2).

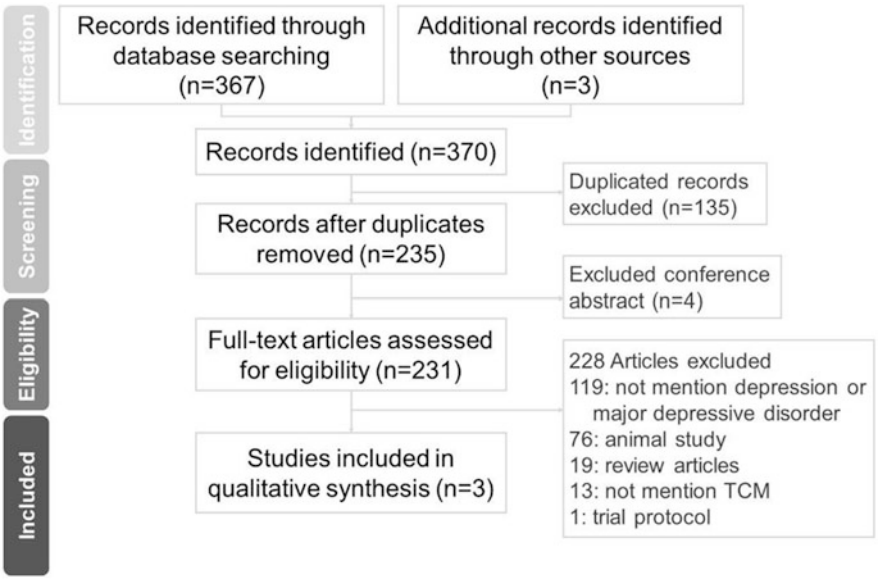


**Fig. 19.1** Preferred reporting items for systematic reviews and meta-analysis (PRISMA) guidelines for searching and listing our flowchart for "Depression"

**Table 19.1** Anti-inflammatory effects of TCM in depressive patients

| First author | Year | Study design | N | Intervention group | Treatment duration | Control group | Clinical markers | Inflammatory markers |
|---|---|---|---|---|---|---|---|---|
| Song Cai [4] | 2009 | RCT | 95 | 1. EA + placebo 2. Sham EA + fluoxetine | 6 weeks | Sham EA + placebo | Similar HDRS, CGI | Both EA and fluoxetine reduced the serum IL-1β levels |
| Roxana D. Vázquez [5] | 2011 | RCT | 42 | Real acupuncture | 6 weeks | Sham acupuncture | Lower Carroll rating scale, SCL-90 | Reduced salivary cortisol level |
| Liu Yi [6] | 2015 | RCT | 126 | Acupuncture +SSRI | 6 weeks | SSRI only | Lower MADRS, SERS scores | Lower IL-6, higher IL-4 and IL-10 |

*RCT* randomized controlled trial, *EA* electro-acupuncture, *SSRIs* selective serotonin reuptake inhibitors, *HDRS* hamilton depression rating scale, *CGI* clinical global impression, *SCL-90* symptom checklist-90, *MADRS* montgomery-Asberg depression rating scale, *SERS* side effect rating scale, *IL* interleukin

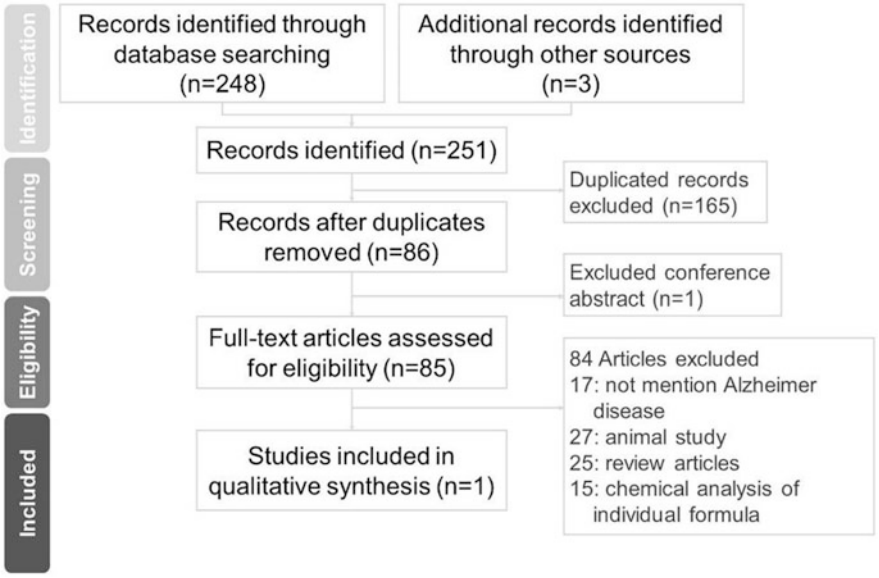


**Fig. 19.2** Preferred reporting items for systematic reviews and meta-analysis (PRISMA) guidelines for searching and listing our flowchart for Alzheimer's disease

### 19.3.3 Parkinson's Disease

We used Preferred Reporting Items for Systematic reviews and Meta-Analysis (PRISMA) guidelines for searching and listed our flowchart (Fig. 19.3). A total of 145 records were identified. Then we excluded duplicated records, articles that didn't mention Parkinson's disease, animal studies, review articles, and articles that didn't mention TCM. We included the studies containing inflammatory markers in Parkinson's disease patients (Table 19.3).

## 19.4 Discussion

### 19.4.1 Depression

Depression is a major psychiatric disease and a leading cause of disability globally [9]. Several researchers devoted their life to the pathogenesis of depression. The monoamine hypothesis had been widely accepted over two decades [10]. It states that the insufficiency of serotonin, norepinephrine, or dopamine in the central nervous system contributed to depression. Then, the mainstay antidepressive drugs were SSRIs (selective serotonin reuptake inhibitors) [11]. In addition, the neuroinflammation hypothesis had been developed to explain the cause of depression [12]. The clinical investigations found elevated pro-inflammatory cytokines levels in depressive patients, including IL-6 [13–20], IL-1β [15], and TNF [14, 16,

**Table 19.2** Anti-inflammatory effects of TCM in Alzheimer's disease patients

| First author | Year | Study design | N | Intervention group | Treatment duration | Control group | Clinical markers | Inflammatory markers |
|---|---|---|---|---|---|---|---|---|
| Hong-Lin Chen [7] | 2016 | RCT | 200 | Huanglian Jiedu decoction TID | 12 weeks | Pitavastatin 2 mg po QD | No | After treatment, the expression levels of IL-1β, IL-6, and TNF were reduced in both groups. And the intervention group levels were decreased more than the control group |

*RCT* randomized controlled trial, *IL* interleukin, *TNF* tumor necrosis factor

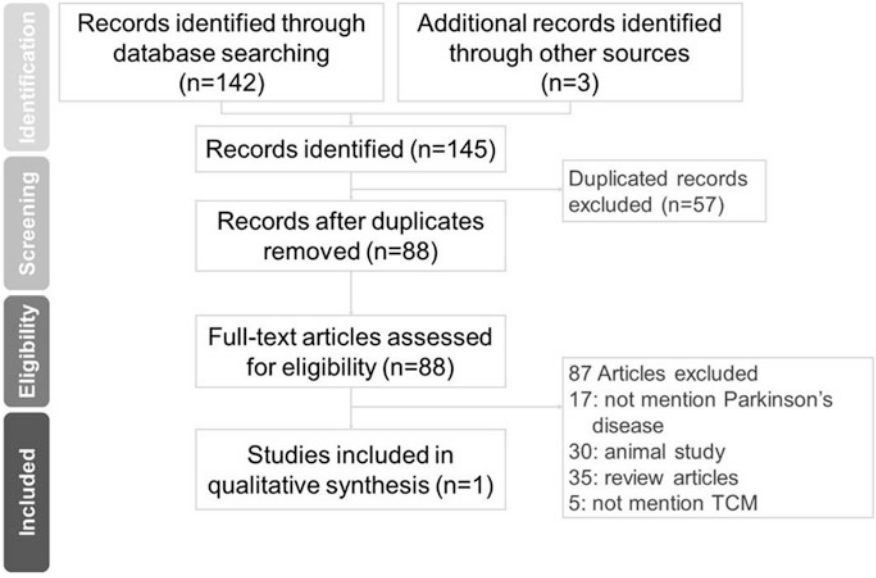


**Fig. 19.3** Preferred reporting items for systematic reviews and meta-analysis (PRISMA) guidelines for searching and listing our flowchart for Parkinson's disease

20, 21], with reduced anti-inflammatory cytokines, including IL-10 [22] and IL-13 [23]. On the other hand, the patients who received interferon-α developed behavioral changes, such as depressive mood, anxiety, and cognitive impairment [24]. The study using functional imaging 18F positron-emission tomography (PET) demonstrated that interferon-α injection affected the activity of the prefrontal cortex and basal ganglia, which are associated with depressive symptoms [25, 26]. The cytokines had interaction with monoamines, particularly serotonin [12]. Several findings reported that cytokines (IL-1, IL-6, interferon-α) [27] increase hypothalamic-pituitary-adrenal (HPA) axis activity by increasing mRNA and the protein of corticotropin-releasing hormone (CRH), thus contributing to the onset of depression in the animal model [28]. The depressive patients showed cognitive impairment and reduced hippocampal volume by weakening neurogenesis [29, 30].

Furthermore, clinical studies reported that autoimmune diseases such as multiple sclerosis [31], rheumatoid arthritis [32], or systemic lupus erythematosus [33] are associated with depression [34]. Anti-inflammatory drugs like celecoxib, aspirin, minocycline, omega-3 fatty acids, and neurosteroids are used as supplementary treatment for depression [35–43].

Despite the widespread use of SSRIs, only 27% of depression patients got remission clinically after a 14-week persistent and vigorous treatment regime [44, 45]. After 1 year of treatment, including drug and nondrug intervention, one-third of depressive patients remained significantly ill [46, 47]. Because of this unmet medical need (enduring impairments in function and persistence of symptoms) [48], many depressive patients seek alternative therapy. In the United

**Table 19.3** Anti-inflammatory effects of electro-acupuncture in Parkinson's disease patients

| First author | Year | Study design | *N* | Intervention group | Treatment duration | Control group | Clinical markers | Inflammatory markers |
|---|---|---|---|---|---|---|---|---|
| Fang Wang [8] | 2015 | RCT | 48 | Drug+EA | Q3D for 2 months | Drug alone | Reduced UPDRS II and III scores, HDRS, and Pittsburgh sleep quality index | Slow down the increase of nitric oxide level in serum (compared to drug alone group) |

*RCT* randomized controlled trial, *EA* electro-acupuncture, *Q3D* one time of acupuncture per 3 days, *UPDRS* unified parkinson's disease rating scale, *HDRS* hamilton depression rating scale

States, 53.6% of depressive people use some forms of complementary and alternative (CAM) therapies to deal with their depression [49].

TCM is one of the oldest medical treatments in the world, and it is a common form of complementary and alternative medicine (CAM) therapy for depression [50, 51]. The onset of depression is often due to "damage" caused by extreme emotion. TCM is based on individual patients' patterns of diagnosis [2]. TCM pattern differentiation is a diagnostic conclusion of the pathological changes of a disease state based on an individual's symptoms, physical signs, pulse form, and tongue appearance [52]. There are eight major parameters, yin (陰) and yang (陽), external (表) and internal (裡), cold (寒) and hot (熱), and the deficiency (虛) and excess (實), that describe the patterns of bodily disharmony. Additional systems, such as qi (氣), blood (血), and body fluid (津液) differentiation and zang-fu (臟腑) (organ) differentiation are also used [2]. In 2015, a research team in Hong Kong performed a comprehensive systemic review of Chinese herbal medicine treatment for depression, including 61 studies, 2504 subjects, and 27 TCM patterns [52]. They found the top four commonly studied TCM patterns were liver qi depression (肝氣鬱結), liver depression and spleen deficiency (肝鬱脾虛), dual deficiency of the heart and spleen (心脾兩虛), and liver depression and qi stagnation (肝鬱氣滯). Bai Shao (*Paeonia lactiflora Pall.*) (白芍) and Chai Hu (*Bupleurum chinense* DC.) (柴胡) were most commonly used across different TCM patterns regardless of the prescribed Chinese herbal formulae. Bai Shao had the function of nourishing the blood and emolliating the liver. In an animal study, the antidepressant effect of Bai Shao may be through the modulation of the function of the hypothalamic-pituitary-adrenal axis [53]. Chai Hu, which can soothe the liver, was found to have hepatoprotective, anti-inflammatory, antipyretic, analgesic, and immunomodulatory effects [54, 55].

Acupuncture is an important component of TCM. Previous research demonstrated the anti-inflammatory effect of acupuncture in depressive disorder [56, 57]. In 2018, our team published a systemic review [2] of the effectiveness of acupuncture combined with manual, laser, or electro-acupuncture in treating depression. We investigated 26 acupuncture-based randomized control trials (RCTs) involving 2618 participants. In most head-to-head clinical trials, acupuncture and medication did not differ significantly in reducing depressive symptoms. But acupuncture might be more effective than medication in reducing specific symptom clusters such as anxiety, somatization and cognitive disturbance [58], and faster onset than antidepressant drugs [59]. However, these findings should be interpreted with caution because they were not conducted under double-blind conditions. A few studies [60, 61] comparing active and inactive laser acupuncture demonstrated a definite superiority with active treatment, eliminating the placebo effect associated with acupuncture. Finally, electro-acupuncture was associated with positive antidepressant results [62, 63].

The above evidence convinced us that TCM had an antidepressive and anti-inflammatory effect. We are curious about human clinical studies of the antidepressive effect of TCM via the anti-inflammatory mechanism. Therefore, we conducted this systemic review.

Although many animal studies demonstrated the anti-inflammatory effect of TCM in depression [64–67], only three human clinical studies investigated the anti-inflammatory effect in depressive patients. In 2009, Song et al. compared 95 depressive patients and 30 healthy controls. They found that the depressive patients had a higher pro-inflammatory cytokine, interleukin (IL)-1β, and a lower anti-inflammatory cytokine, IL-10 [4]. Furthermore, they divided the 95 depressive patients into 3 groups: electro-acupuncture and placebo capsules, sham electro-acupuncture and fluoxetine, and sham electro-acupuncture and placebo capsules. Then, both electro-acupuncture and fluoxetine treatments, but not the placebo, reduced IL-1β concentrations in responders. However, only electro-acupuncture attenuated TNF-α concentration and INF-γ/IL-4 ratio compared with the control group. This was the first randomized controlled trial supporting the anti-inflammatory effect of electro-acupuncture (by restoring the balance between Th1 and Th2 systems via increasing TNF-α and decreasing IL-4).

In 2011, a Mexican group recruited 42 depressive patients and divided them into real acupuncture and sham acupuncture groups [5]. The real acupuncture treatment reduced the depressive symptoms (by Carroll rating scale and SCL-90) and the salivary cortisol level. This study pointed out the anti-inflammatory effect of real electro-acupuncture in depression and the mechanism involved the hypothalamic-pituitary-adrenal (HPA) axis.

In 2015, a research team in Hangzhou, China, published an article on smoothing-liver and nourishing-heart acupuncture for depression [6]. They divided the 126 participants into the SSRI group (65 patients) and SSRI-added acupuncture group (61 patients). They found that the acupuncture add-on group had lower MADRS and SERS scores at weeks 1, 2, 4, and 6 after treatment. And the cytokine analysis found lower IL-6 in the acupuncture add-on group. The anti-inflammatory cytokines IL-4 and IL-10 were significantly higher in the acupuncture add-on group. The authors stated that acupuncture could regulate the balance of pro-inflammatory cytokines and anti-inflammatory cytokines, which was compatible with the findings in the animal models [57, 68].

### 19.4.2 Alzheimer's Disease

AD is the most common neurodegenerative disease worldwide [69]. The percentage of people with AD increases dramatically with age [70]. The death resulting from stroke, HIV, and heart disease has decreased, whereas death from AD is increased to 146.2% [70]. But most of the current treatments failed in combating AD [71]. There are significant unmet medical needs among AD patients. Many patients tried to seek alternative treatment to alleviate their symptoms. In Taiwan, among the 1137 newly diagnosed AD patients, between 1997 and 2008, 78.2% used TCM treatments, including Chinese herbal remedies and acupuncture [72].

In TCM theory, the brain is an outgrowth of and is nourished by the kidney [73]. The energy from the kidney, called kidney essence, can produce marrow, including cerebral marrow, spinal cord, and bone marrow. As Huangdi's Classics

on Medicine [74] (黃帝內經) said: "the brain is sea of marrow," and "kidney stores essence to generate marrow." Therefore the kidney essence deficiency relates to AD. Other studies stated that AD might be caused by spleen deficiency, qi and blood deficiency, or blood stasis in collaterals [75, 76]. Many clinical and basic studies demonstrate the evidence of TCM (including acupuncture) in treating AD.

First, we briefly summarized the clinical evidence of using TCM to treat AD. The single herbs and herbal formulae are used in treating AD [77]. In 2012, a premodern literature review organized 127 Chinese medical books and identified 31 herbs used in treating dementia [78]. Of the 110 different natural products identified, the most frequently cited for dementia were yuan zhi (*Polygala tenuifolia*), fushen (*Poria cocos*), and changpu (*Acorus* spp.). A nationwide, population-based cohort study conducted in Taiwan [72] found that the Bu-Zhong-Yi-Qi-Tang and Ji-Sheng-Shen-Qi-Wan were the two formulae most frequently prescribed by TCM practitioners to treat AD. The female patients living in urban areas were more likely to use Chinese medicine to treat AD [72]. Another study found young-onset dementia, a higher number of BPSD, multiple chronic diseases, and polypharmacy were independent predictors for dementia patients seeking TCM medical advice [79]. An integrated study found benzodiazepines (BZD) were the most common sedative drugs combined with traditional Chinese formulae. Both neurologists and TCM practitioners focused on treating the sleep problems of dementia patients and on a significant number of co-prescriptions of hypnotic drugs and sedative herbal formulas [80]. Furthermore, a multicenter, randomized, double-blind trial showed the effect of Jia-Wei-Xiao-Yao-San in reducing depression and anxiety, which commonly occur in the course of AD [81].

One of the important mechanisms of these formulas was anti-inflammation. For example, the major component of the frequently prescribed TCM (Tian-Ma-Gou-Teng-Yin, Ban-Xia-Xie-Xin-Tang, and Chai-Hu-Jia-Long-Gu-Mu-Li-Tang) was *Scutellaria baicalensis* [80]. In addition, some studies showed its sleep-inducing and sedative effect [82]. In animal studies, there were numerous TCM formulas that had an anti-inflammatory or immunomodulatory effect [83]. The ginkgo biloba extract EGb 761 displayed anti-neuroinflammatory activity, reduced neuroinflammatory activation by targeting the COX/PGE2 pathway [84].

Acupuncture is a widely used non-pharmacologic therapy to treat physical pain [85, 86], and it is also effective in treating AD [87]. Clinically, there were some systemic reviews and meta-analyses that demonstrated the treatment effect of acupuncture in AD, including acupuncture [87], acupuncture plus herbal medicine [88], and acupuncture plus western drugs [89].

Acupuncture is documented to regulate the Aβ metabolism, tau phosphorylation, neurotransmitters, neurogenesis, etc. [90]. In Senescence-Accelerated Prone 8 (SAM-P8) mice, the acupuncture treatment inhibited the PI3K/PDK1/Npkc/Rac1 signaling inflammatory pathway and improved cognitive function [91]. Acupuncture is said to downregulate the NLRP3 inflammasome and decrease the production of downstream pro-inflammatory cytokines like IL-1β and Caspase-1, thus improving cognitive function [92, 93]. Another Aβ-induced AD rat model found the anti-inflammatory effect of electro-acupuncture by downregulating the JAK/STAT3

pathway [94]. Cai et al. used the 5xFAD mice model and found EA stimulation ameliorated cognitive impairment by inhibiting neuroinflammation. Then, the CD11b (for microglia) and GFAP (for astrocytes) expression in the prefrontal cortex of mice was decreased [95].

The above evidence demonstrated that TCM, including acupuncture, could ameliorate the cognitive impairment in AD. Then, we conducted a systemic review of the clinical study of TCM in AD via an anti-inflammatory mechanism.

By our systemic review, we found an article published in 2016, showing the effect of Huang-lian Jie-du decoction (composed of *Coptis*, *Scutellaria*, *Phellodendron*, and *Gardenia*, belonging to "Qing Re Yao" [96]) in treating AD [7]. The authors conducted a randomized controlled trial and found that Huang-lian Jie-du decoction reduced the level of pro-inflammatory cytokines, such as IL-1β, IL-6, and TNF-α in blood, and Aβ-1-42 protein, phosphorylated tau protein in cerebrospinal fluid. As a result, the authors suggested the anti-inflammatory effect of Huang-lian Jie-du decoction in AD patients.

Previous research had discovered the cerebrospinal fluid biomarkers in AD, such as Aβ-1-42 and phosphorylated tau protein [97]. Researchers have proposed that the pro-inflammatory cytokines promote the production of Aβ peptides [98]. These inflammatory cytokines, such as IL-1β, tumor necrosis factor-α (TNF-α), and IL-6, induced neuroinflammation via deposition of Aβ plaques and thus aggravated neurotoxic effects [99]. In addition, elevated levels of pro-inflammatory cytokines in the cerebrospinal fluid and blood in AD patients were observed [100–103].

### 19.4.3 Parkinson's Disease

PD is the second most common neurodegenerative disease, complex and generally progressive, with many motor and non-motor symptoms [104]. In 2020, an article published on "Nature Review" comprehensively analyzed evidence for immune system involvement in PD [105]. The clinical observation studies found an increased risk of PD in patients with autoimmune disease [106–113]. The anti-inflammatory drugs or immunosuppressants are associated with a lower risk of PD [114, 115]. In 1988, the postmortem PD brain specimen analysis found MHC class II molecules, indicating the involvement of neuroinflammation in PD [116]. Pro-inflammatory cytokines (IL-1β, IL-2, IL-6, and TNF) are present at high levels in the CSF of patients with PD and the striatum of postmortem brains from PD patients [117–120]. The high levels of IL-6 in peripheral blood or CSF had been associated with an increased risk of PD [121–123]. In addition, the cellular immune response was also involved in PD patients. A study that compared the peripheral blood of PD patients and healthy controls showed that monocytes and their precursors are upregulated in PD patients [124]. In a mouse model, the peripheral monocyte upregulation was associated with the expression of full-length α-synuclein [125]. As we know, α-synuclein is a presynaptic neuronal protein linked genetically and neuropathologically to PD [126]. In addition, monocytes isolated from PD patients expressed a higher level of the PD-associated protein, LRRK2, than those from

healthy controls [127]. Several findings found that impaired lysosomal function in monocytes leads to failure of α-synuclein clearance and contributes to PD [128]. PD patients had altered gut microbiota and inflammatory markers in the feces [129]. The brain image reveals activated microglia in the PD patients [130–132]. Bioinformatic analysis showed that PD and autoimmune diseases share similar molecular pathways and polygenic risk variants [133–135]. Then, the genetic analysis revealed that some loci associated with PD are associated with the immune function [136, 137].

Current modern treatment for PD is symptomatic; no disease-modifying pharmacologic therapies are available [138]. Most neurologists use dopamine-based therapies to treat motor symptoms and nondopaminergic approaches (SSRIs for psychiatric symptoms) to treat nonmotor symptoms—the advanced treatment being deep brain stimulation, levodopa-carbidopa enteral suspension for medication-resistant tremors or dyskinesias. But some patients still got an unsatisfactory response to the above treatment [139], explaining why the patients chose to receive alternative therapy for treating PD. 40% of PD patients visited alternative therapy providers for their symptoms [140].

TCM is an important alternative therapy widely used globally, especially in East Asia [141]. James Parkinson originally described PD in 1817 [142]; therefore, the term "Parkinson" could not be found in ancient TCM books. But centuries ago, the TCM bible, Huangdi Neijing (Huangdi's Internal Classic) [143], described a single herb or herbal formula in treating similar disorders, featured by tremor, rigidity, bradykinesia, and gait disturbance, currently considered as PD [144]. In TCM, PD corresponds to Chan Zheng (tremor), Chan Zhen (shaking), and Dong Feng (wind stirring). In a systemic database review over the last 30 years, the authors documented PD's most frequent TCM patterns. The top five were "Yin deficiency of kidney and liver," "a deficiency of Qi and blood," "phlegm heat and wind stirring," "blood stasis and wind stirring," and "a deficiency of Yin and Yang" [145].

In 2017, Li et al. published a systemic review of the mechanism of TCM in treating PD [144]. TCM could inhibit oxidative stress in the central nervous system, regulate mitochondrial dysfunction, inhibit neuronal apoptosis, inhibit abnormal protein aggregation, inhibit neuroinflammation, etc. Curcumin had the effect to protect dopaminergic neurons [146], and celastrol could delay the progression of PD [147].

Acupuncture is an important component of TCM. In 2005, a double-blind pilot study found that the patients who received acupuncture had nonsignificant trends toward improvement in the activities of daily living score of the PDQ-39 [148]. Later, there were rising numbers of clinical studies investigating the effect of acupuncture in treating PD. In 2020, a study conducted a qualitative assessment of 11 systemic reviews or meta-analyses of acupuncture in PD [149]. They found that 10 of the 11 systemic reviews/meta-analyses reached positive effect (90.9%). But most studies didn't draw firm conclusions about acupuncture due to small sample sizes or low methodological quality. The authors stated that acupuncture might be a promising treatment for PD, especially for motor symptoms. But additional studies with rigorous experimental designs and larger sample sizes are needed to verify these results.

In respect to the mechanism of acupuncture in PD, a few animal studies demonstrated the reduction of oxidative stress and neuroinflammation [150] by inhibiting microglial activation [151], stimulating the release of neurotrophic factors, or regulating the network between the cortex and striatum [152]. Here we mainly discussed the neuroinflammation and gut-brain axis.

Recent evidence demonstrated that the gut-brain interaction is relevant to the pathogenesis of PD [153, 154]. PD patients are known to exhibit constipation, which is currently known as a prodromal symptom of PD. The evidence supports the Braak hypothesis that α-synuclein pathology spreads from the intestine to the brainstem via the vagus nerve and then ascends to the substantia nigra [155–158]. The inflammatory process in the gut plays a vital role in the pathogenesis of PD [159–161]. In a clinical study, PD patients had higher pro-inflammatory cytokines and colon glial cell activation levels than healthy controls [162]. In addition, PD patients had altered gut microbiota composition than healthy controls [163]. The alteration of gut microbiota was associated with plasma cytokines changes. Furthermore, the stool samples in PD patients had higher levels of TLR4, T cells, and cytokines than those in healthy controls [129].

Acupuncture had shown the effect of neuroprotection, anti-inflammation, and anti-apoptosis in PD mice models [164–170]. In the mice model of sepsis, acupuncture could regulate the immune system, possibly via the vagus nerve, which was widely discussed in the gut-brain axis [171]. The relationship between neuroinflammation and the gut-brain axis was observed in the animal model. Acupuncture is found to alter the gut microbiota and inhibit neuroinflammation in the substantia nigra and striatum. Furthermore, this regulation improved the motor function of mice and protected the dopaminergic neurons [172].

We did a systemic review of acupuncture and TCM in treating PD via anti-inflammatory mechanisms. In a well-designed randomized clinical trial [8], participants were randomized to drug alone ($n = 20$) or drug plus electro-acupuncture (EA) group ($n = 28$). They found clinically better responses in patients with the EA add-on group (by UPDRS, HDRS, and Pittsburgh Sleep Quality Index). The result showed that the EA add-on group had a lower nitric oxide (NO) in serum (compared to the drug alone group). The result was compatible with previous evidence [173] that higher NO levels in PD patients lead to higher UPDRS scores, which means more severe PD symptoms. In summary, this study demonstrated the clinical effectiveness and anti-inflammatory mechanism of EA add-on treatment in PD patients.

In summary, we did a systemic review of the anti-inflammatory effect of TCM, including acupuncture, in treating depression, AD, and PD. Most of the detailed mechanisms were found by basic studies. We organized the anti-inflammatory mechanism and presented it in Fig. 19.4.

The major limitation of TCM application is that the research results have been inconsistent in design quality and clinical powers [1]. With financial incentives from patent protection, pharmaceutical companies are more willing to invest in expensive RCTs with the highest standard methodology to successfully detect the small

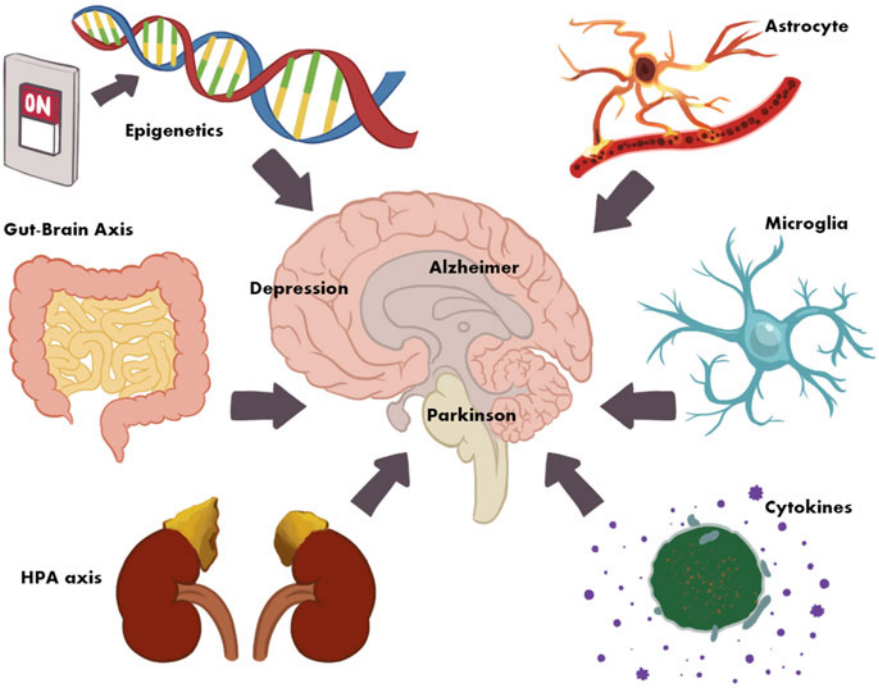


**Fig. 19.4** A brief summary of the anti-inflammatory mechanism of TCM on the concept of mind-body interface. TCM could modulate astrocyte, microglia in the central nervous system. TCM also had an effect on cytokines and HPA axis. TCM could regulate the gut-brain axis and epigenetics. *HPA* hypothalamic-pituitary-adrenal axis

therapeutic effects of complex diseases. However, most of the TCM, including acupuncture, are non-patentable treatments.

## 19.5 Conclusion

Both basic and clinical studies demonstrated the anti-inflammatory and immunomodulatory effects of TCM, including acupuncture, in treating depression, AD, and PD. TCM, including acupuncture, may be introduced to patients with unsatisfactory treatment response by current management. Future research will be needed to investigate the anti-inflammatory effect of TCM further.

## References

1. Su KP. Are we all the same? The critical role of translational brain, behavior, and immunity research in East Asia. Brain Behav Immun. 2019;82:1–2.

2. Su K-P, Chou L-W, Sun M-F, Lin J-G. Acupuncture treatment in depression. In: Lin J-G, editor. Experimental Acupuncturology. Singapore: Springer Singapore; 2018. p. 43–66.
3. NIH Consensus Conference. Acupuncture. JAMA. 1998;280(17):1518–24.
4. Song C, Halbreich U, Han C, Leonard BE, Luo H. Imbalance between pro- and anti-inflammatory cytokines, and between Th1 and Th2 cytokines in depressed patients: the effect of electroacupuncture or fluoxetine treatment. Pharmacopsychiatry. 2009;42(5):182–8.
5. Vázquez RD, González-Macías L, Berlanga C, Aedo FJ. Effect of acupuncture treatment on depression: correlation between psychological outcomes and salivary cortisol levels. Salud mental. 2011;34:21–6.
6. Liu Y, Feng H, Mo Y, Gao J, Mao H, Song M, et al. Effect of soothing-liver and nourishing-heart acupuncture on early selective serotonin reuptake inhibitor treatment onset for depressive disorder and related indicators of neuroimmunology: a randomized controlled clinical trial. J Tradit Chin Med. 2015;35(5):507–13.
7. Chen H-L, Guan F. Effect of Huanglian jiedu decoction on pitavastatin treatment of Alzheimer's disease. Journal of Hainan Medical University. 2016;22:6.
8. Wang F, Sun L, Zhang XZ, Jia J, Liu Z, Huang XY, et al. Effect and potential mechanism of Electroacupuncture add-on treatment in patients with Parkinson's disease. Evid Based Complement Alternat Med. 2015;2015:692795.
9. Chou PH, Lin YF, Lu MK, Chang HA, Chu CS, Chang WH, et al. Personalization of repetitive transcranial magnetic stimulation for the treatment of major depressive disorder according to the existing psychiatric comorbidity. Clin Psychopharmacol Neurosci. 2021;19(2):190–205.
10. Delgado PL. Depression: the case for a monoamine deficiency. J Clin Psychiatry. 2000;61 (Suppl 6):7–11.
11. Clevenger SS, Malhotra D, Dang J, Vanle B, IsHak WW. The role of selective serotonin reuptake inhibitors in preventing relapse of major depressive disorder. Ther Adv Psychopharmacol. 2018;8(1):49–58.
12. Jeon SW, Kim YK. The role of neuroinflammation and neurovascular dysfunction in major depressive disorder. J Inflamm Res. 2018;11:179–92.
13. Alesci S, Martinez PE, Kelkar S, Ilias I, Ronsaville DS, Listwak SJ, et al. Major depression is associated with significant diurnal elevations in plasma Interleukin-6 levels, a shift of its circadian rhythm, and loss of physiological complexity in its secretion: clinical implications. J Clin Endocrinol Metabol. 2005;90(5):2522–30.
14. Dowlati Y, Herrmann N, Swardfager W, Liu H, Sham L, Reim EK, et al. A meta-analysis of cytokines in major depression. Biol Psychiatry. 2010;67(5):446–57.
15. Howren MB, Lamkin DM, Suls J. Associations of depression with C-reactive protein, IL-1, and IL-6: a meta-analysis. Psychosom Med. 2009;71(2):171–86.
16. Lanquillon S, Krieg JC, Bening-Abu-Shach U, Vedder H. Cytokine production and treatment response in major depressive disorder. Neuropsychopharmacology. 2000;22(4):370–9.
17. Miller AH, Maletic V, Raison CL. Inflammation and its discontents: the role of cytokines in the pathophysiology of major depression. Biol Psychiatry. 2009;65(9):732–41.
18. Raison CL, Capuron L, Miller AH. Cytokines sing the blues: inflammation and the pathogenesis of depression. Trends Immunol. 2006;27(1):24–31.
19. Su S, Miller AH, Snieder H, Bremner JD, Ritchie J, Maisano C, et al. Common genetic contributions to depressive symptoms and inflammatory markers in middle-aged men: the twins heart study. Psychosom Med. 2009;71(2):152–8.
20. Yang K, Xie G, Zhang Z, Wang C, Li W, Zhou W, et al. Levels of serum interleukin (IL)-6, IL-1beta, tumour necrosis factor-alpha and leptin and their correlation in depression. Aust N Z J Psychiatry. 2007;41(3):266–73.
21. Tuglu C, Kara SH, Caliyurt O, Vardar E, Abay E. Increased serum tumor necrosis factor-alpha levels and treatment response in major depressive disorder. Psychopharmacology (Berl). 2003;170(4):429–33.
22. Clerici M, Arosio B, Mundo E, Cattaneo E, Pozzoli S, Dell'osso B, et al. Cytokine polymorphisms in the pathophysiology of mood disorders. CNS Spectr. 2009;14(8):419–25.

23. Wong ML, Dong C, Maestre-Mesa J, Licinio J. Polymorphisms in inflammation-related genes are associated with susceptibility to major depression and antidepressant response. Mol Psychiatry. 2008;13(8):800–12.
24. Al-Huthail Y. Neuropsychiatric side-effects of interferon alfa therapy for hepatitis C and their management: a review. Saudi J Gastroenterol. 2006;12(2):59–67.
25. Capuron L, Ravaud A, Dantzer R. Timing and specificity of the cognitive changes induced by interleukin-2 and interferon-alpha treatments in cancer patients. Psychosom Med. 2001;63(3): 376–86.
26. Eisch AJ, Petrik D. Depression and hippocampal neurogenesis: a road to remission? Science. 2012;338(6103):72–5.
27. Pace TW, Hu F, Miller AH. Cytokine-effects on glucocorticoid receptor function: relevance to glucocorticoid resistance and the pathophysiology and treatment of major depression. Brain Behav Immun. 2007;21(1):9–19.
28. De Souza EB. Corticotropin-releasing factor receptors: physiology, pharmacology, biochemistry and role in central nervous system and immune disorders. Psychoneuroendocrinology. 1995;20(8):789–819.
29. Mahar I, Bambico FR, Mechawar N, Nobrega JN. Stress, serotonin, and hippocampal neurogenesis in relation to depression and antidepressant effects. Neurosci Biobehav Rev. 2014;38:173–92.
30. Kim YK, Na KS, Myint AM, Leonard BE. The role of pro-inflammatory cytokines in neuroinflammation, neurogenesis and the neuroendocrine system in major depression. Prog Neuropsychopharmacol Biol Psychiatry. 2016;64:277–84.
31. Rossi S, Studer V, Motta C, Polidoro S, Perugini J, Macchiarulo G, et al. Neuroinflammation drives anxiety and depression in relapsing-remitting multiple sclerosis. Neurology. 2017;89 (13):1338–47.
32. Margaretten M, Julian L, Katz P, Yelin E. Depression in patients with rheumatoid arthritis: description, causes and mechanisms. Int J Clin Rheumtol. 2011;6(6):617–23.
33. Figueiredo-Braga M, Cornaby C, Cortez A, Bernardes M, Terroso G, Figueiredo M, et al. Depression and anxiety in systemic lupus erythematosus: the crosstalk between immunological, clinical, and psychosocial factors. Medicine (Baltimore). 2018;97(28):e11376.
34. Pollak Y, Yirmiya R. Cytokine-induced changes in mood and behaviour: implications for 'depression due to a general medical condition', immunotherapy and antidepressive treatment. Int J Neuropsychopharmacol. 2002;5(4):389–99.
35. Eyre HA, Air T, Proctor S, Rositano S, Baune BT. A critical review of the efficacy of non-steroidal anti-inflammatory drugs in depression. Prog Neuropsychopharmacol Biol Psychiatry. 2015;57:11–6.
36. García-Bueno B, Pérez-Nievas BG, Leza JC. Is there a role for the nuclear receptor PPARγ in neuropsychiatric diseases? Int J Neuropsychopharmacol. 2010;13(10):1411–29.
37. Johansson D, Falk A, Marcus MM, Svensson TH. Celecoxib enhances the effect of reboxetine and fluoxetine on cortical noradrenaline and serotonin output in the rat. Prog Neuropsychopharmacol Biol Psychiatry. 2012;39(1):143–8.
38. Köhler O, Benros ME, Nordentoft M, Farkouh ME, Iyengar RL, Mors O, et al. Effect of anti-inflammatory treatment on depression, depressive symptoms, and adverse effects: a systematic review and meta-analysis of randomized clinical trials. JAMA Psychiat. 2014;71(12): 1381–91.
39. Krebs M, Leopold K, Hinzpeter A, Schaefer M. Neuroprotective agents in schizophrenia and affective disorders. Expert Opin Pharmacother. 2006;7(7):837–48.
40. Müller N, Schwarz MJ, Dehning S, Douhe A, Cerovecki A, Goldstein-Müller B, et al. The cyclooxygenase-2 inhibitor celecoxib has therapeutic effects in major depression: results of a double-blind, randomized, placebo controlled, add-on pilot study to reboxetine. Mol Psychiatry. 2006;11(7):680–4.
41. Najjar S, Pearlman DM, Alper K, Najjar A, Devinsky O. Neuroinflammation and psychiatric illness. J Neuroinflammation. 2013;10:43.

42. Nery FG, Monkul ES, Hatch JP, Fonseca M, Zunta-Soares GB, Frey BN, et al. Celecoxib as an adjunct in the treatment of depressive or mixed episodes of bipolar disorder: a double-blind, randomized, placebo-controlled study. Hum Psychopharmacol. 2008;23(2):87–94.
43. Schmidt FM, Kirkby KC, Himmerich H. The TNF-alpha inhibitor etanercept as monotherapy in treatment-resistant depression—report of two cases. Psychiatr Danub. 2014;26(3):288–90.
44. Rush AJ. STAR*D: what have we learned? Am J Psychiatry. 2007;164(2):201–4.
45. Trivedi MH, Rush AJ, Wisniewski SR, Nierenberg AA, Warden D, Ritz L, et al. Evaluation of outcomes with citalopram for depression using measurement-based care in STAR*D: implications for clinical practice. Am J Psychiatry. 2006;163(1):28–40.
46. Gaynes BN, Rush AJ, Trivedi MH, Wisniewski SR, Spencer D, Fava M. The STAR*D study: treating depression in the real world. Cleve Clin J Med. 2008;75(1):57–66.
47. Sinyor M, Schaffer A, Levitt A. The sequenced treatment alternatives to relieve depression (STAR*D) trial: a review. Can J Psychiatry. 2010;55(3):126–35.
48. Solomon D, Adams J. The use of complementary and alternative medicine in adults with depressive disorders. A critical integrative review. J Affect Disord. 2015;179:101–13.
49. Kessler RC, Soukup J, Davis RB, Foster DF, Wilkey SA, Van Rompay MI, et al. The use of complementary and alternative therapies to treat anxiety and depression in the United States. Am J Psychiatry. 2001;158(2):289–94.
50. Hsu M-C, Creedy D, Moyle W, Venturato L, Tsay S-L, Ouyang W-C. Use of complementary and alternative medicine among adult patients for depression in Taiwan. J Affect Disord. 2008;111(2):360–5.
51. Qureshi NA, Al-Bedah AM. Mood disorders and complementary and alternative medicine: a literature review. Neuropsychiatr Dis Treat. 2013;9:639–58.
52. Yeung WF, Chung KF, Ng KY, Yu YM, Zhang SP, Ng BF, et al. Prescription of Chinese herbal medicine in pattern-based traditional Chinese medicine treatment for depression: a systematic review. Evid Based Complement Alternat Med. 2015;2015:160189.
53. Mao QQ, Ip SP, Xian YF, Hu Z, Che CT. Anti-depressant-like effect of peony: a mini-review. Pharm Biol. 2012;50(1):72–7.
54. Sun HX. Haemolytic activities and adjuvant effect of Bupleurum chinense saponins on the immune responses to ovalbumin in mice. Vaccine. 2006;24(9):1324–31.
55. Yeung WF, Chung KF, Poon MM, Ho FY, Zhang SP, Zhang ZJ, et al. Prescription of chinese herbal medicine and selection of acupoints in pattern-based traditional chinese medicine treatment for insomnia: a systematic review. Evid Based Complement Alternat Med. 2012;2012:902578.
56. Satyanarayanan SK, Shih Y-H, Wen Y-R, Palani M, Lin Y-W, Su H, et al. miR-200a-3p modulates gene expression in comorbid pain and depression: molecular implication for central sensitization. Brain Behav Immun. 2019;82:230–8.
57. Lin Y-W, Chou AIW, Su H, Su K-P. Transient receptor potential V1 (TRPV1) modulates the therapeutic effects for comorbidity of pain and depression: the common molecular implication for electroacupuncture and omega-3 polyunsaturated fatty acids. Brain Behav Immun. 2020;89:604–14.
58. Luo H, Meng F, Jia Y, Zhao X. Clinical research on the therapeutic effect of the electro-acupuncture treatment in patients with depression. Psychiatry Clin Neurosci. 1998;52(Suppl): S338–40.
59. Sun H, Zhao H, Ma C, Bao F, Zhang J, Wang DH, et al. Effects of electroacupuncture on depression and the production of glial cell line-derived neurotrophic factor compared with fluoxetine: a randomized controlled pilot study. J Altern Complement Med. 2013;19(9):733–9.
60. Quah-Smith I, Smith C, Crawford JD, Russell J. Laser acupuncture for depression: a randomised double blind controlled trial using low intensity laser intervention. J Affect Disord. 2013;148(2–3):179–87.
61. Quah-Smith JI, Tang WM, Russell J. Laser acupuncture for mild to moderate depression in a primary care setting—a randomised controlled trial. Acupunct Med. 2005;23(3):103–11.

62. Man SC, Hung BH, Ng RM, Yu XC, Cheung H, Fung MP, et al. A pilot controlled trial of a combination of dense cranial electroacupuncture stimulation and body acupuncture for post-stroke depression. BMC Complement Altern Med. 2014;14:255.
63. Zhang GC, Fu WB, Xu NG, Liu JH, Zhu XP, Liang ZH, et al. Meta analysis of the curative effect of acupuncture on post-stroke depression. J Tradit Chin Med. 2012;32(1):6–11.
64. Chen PJ, Hsieh CL, Su KP, Hou YC, Chiang HM, Lin IH, et al. The antidepressant effect of Gastrodia elata Bl. On the forced-swimming test in rats. Am J Chin Med. 2008;36(1):95–106.
65. Chen PJ, Hsieh CL, Su KP, Hou YC, Chiang HM, Sheen LY. Rhizomes of Gastrodia elata B (L) possess antidepressant-like effect via monoamine modulation in subchronic animal model. Am J Chin Med. 2009;37(6):1113–24.
66. Chen PJ, Liang KC, Lin HC, Hsieh CL, Su KP, Hung MC, et al. Gastrodia elata Bl. Attenuated learning deficits induced by forced-swimming stress in the inhibitory avoidance task and Morris water maze. J Med Food. 2011;14(6):610–7.
67. Lin SH, Chang HC, Chen PJ, Hsieh CL, Su KP, Sheen LY. The antidepressant-like effect of ethanol extract of daylily flowers (Jīn Zhēn Huā) in rats. J Tradit Complement Med. 2013;3(1): 53–61.
68. Liao HY, Lin YW. Electroacupuncture attenuates chronic inflammatory pain and depression comorbidity through transient receptor potential V1 in the brain. Am J Chin Med. 2021;49(6): 1417–35.
69. Santiago JA, Potashkin JA. The impact of disease comorbidities in Alzheimer's disease. Front Aging Neurosci. 2021;13(38):631770.
70. 2020 Alzheimer's disease facts and figures. Alzheimers Dement. 2020;16(3):391–460.
71. Levey AI. Progress with treatments for Alzheimer's disease. N Engl J Med. 2021;384(18): 1762–3.
72. Lin SK, Yan SH, Lai JN, Tsai TH. Patterns of Chinese medicine use in prescriptions for treating Alzheimer's disease in Taiwan. Chinas Med. 2016;11:12.
73. Liu P, Kong M, Yuan S, Liu J, Wang P. History and experience: a survey of traditional chinese medicine treatment for Alzheimer's disease. Evid Based Complement Alternat Med. 2014;2014:642128.
74. Cavalieri S, Rotoli M. Huangdi Neijing: a classic book of traditional Chinese medicine. Recenti Prog Med. 1997;88(11):541–6.
75. Chen YG. Research Progress in the pathogenesis of Alzheimer's disease. Chin Med J (Engl). 2018;131(13):1618–24.
76. Yu B, Zhou C, Zhang J, Ling Y, Hu Q, Wang Y, et al. Latest study on the relationship between pathological process of inflammatory injury and the syndrome of spleen deficiency and fluid retention in Alzheimer's disease. Evid Based Complement Alternat Med. 2014;2014:743541.
77. Jeon SG, Song EJ, Lee D, Park J, Nam Y, Kim JI, et al. Traditional oriental medicines and Alzheimer's disease. Aging Dis. 2019;10(2):307–28.
78. May BH, Lu C, Lu Y, Zhang AL, Xue CC. Chinese herbs for memory disorders: a review and systematic analysis of classical herbal literature. J Acupunct Meridian Stud. 2013;6(1):2–11.
79. Lin SK, Tsai YT, Lai JN, Wu CT. Demographic and medication characteristics of traditional Chinese medicine users among dementia patients in Taiwan: a nationwide database study. J Ethnopharmacol. 2015;161:108–15.
80. Lin SK, Tzeng JN, Lai JN. The core pattern of Chinese herbal formulae and drug-herb concurrent usage in patients with dementia. Medicine (Baltimore). 2019;98(4):e13931.
81. Park DM, Kim SH, Park YC, Kang WC, Lee SR, Jung IC. The comparative clinical study of efficacy of Gamisoyo-San (Jiaweixiaoyaosan) on generalized anxiety disorder according to differently manufactured preparations: multicenter, randomized, double blind, placebo controlled trial. J Ethnopharmacol. 2014;158(Pt A):11–7.
82. Chang H-H, Yi P-L, Cheng C-H, Lu C-Y, Hsiao Y-T, Tsai Y-F, et al. Biphasic effects of baicalin, an active constituent of Scutellaria baicalensis Georgi, in the spontaneous sleep–wake regulation. J Ethnopharmacol. 2011;135(2):359–68.

83. Zhang Z, Zhang S, Lui CN, Zhu P, Zhang Z, Lin K, Dai Y, Yung KK. Traditional Chinese medicine-based neurorestorative therapy for Alzheimer's and Parkinson's disease. J Neurorestoratology. 2019;7(4):207–22.
84. Gargouri B, Carstensen J, Bhatia HS, Huell M, Dietz GPH, Fiebich BL. Anti-neuroinflammatory effects of Ginkgo biloba extract EGb761 in LPS-activated primary microglial cells. Phytomedicine. 2018;44:45–55.
85. Tsai S-T, Huang W-S, Jiang S-K, Liao H-Y. Cervical spinal epidural abscess following needle-knife acupotomy, with an initial presentation that mimicked an acute stroke: a case report. Hong Kong Journal of Emergency Medicine. 2018;27(2):99–102.
86. Tsai ST, Tseng CH, Lin MC, Liao HY, Teoh BK, San S, et al. Acupuncture reduced the medical expenditure in migraine patients: real-world data of a 10-year national cohort study. Medicine (Baltimore). 2020;99(32):e21345.
87. Huang Q, Luo D, Chen L, Liang FX, Chen R. Effectiveness of acupuncture for Alzheimer's disease: an updated systematic review and meta-analysis. Curr Med Sci. 2019;39(3):500–11.
88. Zhou S, Dong L, He Y, Xiao H. Acupuncture plus herbal medicine for Alzheimer's disease: a systematic review and meta-analysis. Am J Chin Med. 2017;45(7):1327–44.
89. Wang YY, Yu SF, Xue HY, Li Y, Zhao C, Jin YH. Effectiveness and safety of acupuncture for the treatment of Alzheimer's disease: a systematic review and meta-analysis. Front Aging Neurosci. 2020;12:98.
90. Yu CC, Du YJ, Wang SQ, Liu LB, Shen F, Wang L, et al. Experimental evidence of the benefits of acupuncture for Alzheimer's disease: an updated review. Front Neurosci. 2020;14:549772.
91. Li G, Zeng L, Cheng H, Han J, Zhang X, Xie H. Acupuncture administration improves cognitive functions and alleviates inflammation and nuclear damage by regulating phosphatidylinositol 3 kinase (PI3K)/Phosphoinositol-dependent kinase 1 (PDK1)/novel protein kinase C (nPKC)/Rac 1 signaling pathway in senescence-accelerated prone 8 (SAM-P8) mice. Med Sci Monit. 2019;25:4082–93.
92. Ding N, Jiang J, Xu A, Tang Y, Li Z. Manual acupuncture regulates behavior and cerebral blood flow in the SAMP8 mouse model of Alzheimer's disease. Front Neurosci. 2019;13:37.
93. Jiang J, Ding N, Wang K, Li Z. Electroacupuncture could influence the expression of IL-1β and NLRP3 Inflammasome in hippocampus of Alzheimer's disease animal model. Evid Based Complement Alternat Med. 2018;2018:8296824.
94. Tang S-H, Du Y-J, Xiao J-H, Wang Y, Shen F, Sun G-J. Acupuncture and Moxibustion improves learning-memory ability of Alzheimer's disease rats possibly by up-regulating serum Aβ internalization enzyme contents. Zhen Ci Yan Jiu. 2018;43:692–7.
95. Cai M, Lee J-H, Yang EJ. Electroacupuncture attenuates cognition impairment via anti-neuroinflammation in an Alzheimer's disease animal model. J Neuroinflammation. 2019;16(1):264.
96. Guan F, Lam W, Hu R, Kim YK, Han H, Cheng Y-C. Majority of Chinese medicine herb category "Qing re Yao" have multiple mechanisms of anti-inflammatory activity. Sci Rep. 2018;8(1):7416.
97. Niemantsverdriet E, Valckx S, Bjerke M, Engelborghs S. Alzheimer's disease CSF biomarkers: clinical indications and rational use. Acta Neurol Belg. 2017;117(3):591–602.
98. Tuppo EE, Arias HR. The role of inflammation in Alzheimer's disease. Int J Biochem Cell Biol. 2005;37(2):289–305.
99. Belkhelfa M, Rafa H, Medjeber O, Arroul-Lammali A, Behairi N, Abada-Bendib M, et al. IFN-γ and TNF-α are involved during Alzheimer disease progression and correlate with nitric oxide production: a study in Algerian patients. J Interferon Cytokine Res. 2014;34(11):839–47.
100. Brosseron F, Krauthausen M, Kummer M, Heneka MT. Body fluid cytokine levels in mild cognitive impairment and Alzheimer's disease: a comparative overview. Mol Neurobiol. 2014;50(2):534–44.

101. Liu C, Cui G, Zhu M, Kang X, Guo H. Neuroinflammation in Alzheimer's disease: chemokines produced by astrocytes and chemokine receptors. Int J Clin Exp Pathol. 2014;7 (12):8342–55.
102. Rubio-Perez JM, Morillas-Ruiz JM. A review: inflammatory process in Alzheimer's disease, role of cytokines. ScientificWorldJournal. 2012;2012:756357.
103. Swardfager W, Lanctôt K, Rothenburg L, Wong A, Cappell J, Herrmann N. A meta-analysis of cytokines in Alzheimer's disease. Biol Psychiatry. 2010;68(10):930–41.
104. Chiou SM, Lin YC, Lu MK, Tsai CH. Bilateral subthalamic stimulation for advanced Parkinson disease: early experience at an eastern center. Neurol Sci. 2015;36(4):515–20.
105. Tan EK, Chao YX, West A, Chan LL, Poewe W, Jankovic J. Parkinson disease and the immune system—associations, mechanisms and therapeutics. Nat Rev Neurol. 2020;16(6): 303–18.
106. Amanat M, Salehi M, Rezaei N. Neurological and psychiatric disorders in psoriasis. Rev Neurosci. 2018;29(7):805–13.
107. Chang CC, Lin TM, Chang YS, Chen WS, Sheu JJ, Chen YH, et al. Autoimmune rheumatic diseases and the risk of Parkinson disease: a nationwide population-based cohort study in Taiwan. Ann Med. 2018;50(1):83–90.
108. Ju UH, Liu FC, Lin CS, Huang WY, Lin TY, Shen CH, et al. Risk of Parkinson disease in Sjögren syndrome administered ineffective immunosuppressant therapies: a nationwide population-based study. Medicine (Baltimore). 2019;98(14):e14984.
109. Lee JH, Han K, Gee HY. The incidence rates and risk factors of Parkinson disease in patients with psoriasis: a nationwide population-based cohort study. J Am Acad Dermatol. 2020;83(6): 1688–95.
110. Li X, Sundquist J, Sundquist K. Subsequent risks of Parkinson disease in patients with autoimmune and related disorders: a nationwide epidemiological study from Sweden. Neurodegener Dis. 2012;10(1–4):277–84.
111. Rugbjerg K, Friis S, Ritz B, Schernhammer ES, Korbo L, Olsen JH. Autoimmune disease and risk for Parkinson disease: a population-based case-control study. Neurology. 2009;73(18): 1462–8.
112. Sheu JJ, Wang KH, Lin HC, Huang CC. Psoriasis is associated with an increased risk of parkinsonism: a population-based 5-year follow-up study. J Am Acad Dermatol. 2013;68(6): 992–9.
113. Wu MC, Xu X, Chen SM, Tyan YS, Chiou JY, Wang YH, et al. Impact of Sjogren's syndrome on Parkinson's disease: a nationwide case-control study. PLoS One. 2017;12(7):e0175836.
114. Racette BA, Gross A, Vouri SM, Camacho-Soto A, Willis AW, Searles NS. Immunosuppressants and risk of Parkinson disease. Ann Clin Transl Neurol. 2018;5(7): 870–5.
115. Ren L, Yi J, Yang J, Li P, Cheng X, Mao P. Nonsteroidal anti-inflammatory drugs use and risk of Parkinson disease: a dose-response meta-analysis. Medicine (Baltimore). 2018;97(37): e12172.
116. McGeer PL, Itagaki S, McGeer EG. Expression of the histocompatibility glycoprotein HLA-DR in neurological disease. Acta Neuropathol. 1988;76(6):550–7.
117. Karpenko MN, Vasilishina AA, Gromova EA, Muruzheva ZM, Miliukhina IV, Bernadotte A. Interleukin-1β, interleukin-1 receptor antagonist, interleukin-6, interleukin-10, and tumor necrosis factor-α levels in CSF and serum in relation to the clinical diversity of Parkinson's disease. Cell Immunol. 2018;327:77–82.
118. Mogi M, Harada M, Narabayashi H, Inagaki H, Minami M, Nagatsu T. Interleukin (IL)-1 beta, IL-2, IL-4, IL-6 and transforming growth factor-alpha levels are elevated in ventricular cerebrospinal fluid in juvenile parkinsonism and Parkinson's disease. Neurosci Lett. 1996;211(1):13–6.
119. Mogi M, Harada M, Riederer P, Narabayashi H, Fujita K, Nagatsu T. Tumor necrosis factor-alpha (TNF-alpha) increases both in the brain and in the cerebrospinal fluid from Parkinsonian patients. Neurosci Lett. 1994;165(1–2):208–10.

120. Nagatsu T, Mogi M, Ichinose H, Togari A. Cytokines in Parkinson's disease. J Neural Transm Suppl. 2000;58:143–51.
121. Chen H, O'Reilly EJ, Schwarzschild MA, Ascherio A. Peripheral inflammatory biomarkers and risk of Parkinson's disease. Am J Epidemiol. 2008;167(1):90–5.
122. Lian TH, Guo P, Zuo LJ, Hu Y, Yu SY, Yu QJ, et al. Tremor-dominant in Parkinson disease: the relevance to iron metabolism and inflammation. Front Neurosci. 2019;13:255.
123. Schröder JB, Pawlowski M, Meyer Zu Hörste G, Gross CC, Wiendl H, Meuth SG, et al. Immune cell activation in the cerebrospinal fluid of patients with Parkinson's disease. Front Neurol. 2018;9:1081.
124. Wijeyekoon RS, Kronenberg-Versteeg D, Scott KM, Hayat S, Jones JL, Clatworthy MR, et al. Monocyte function in Parkinson's disease and the impact of autologous serum on phagocytosis. Front Neurol. 2018;9:870.
125. Harms AS, Thome AD, Yan Z, Schonhoff AM, Williams GP, Li X, et al. Peripheral monocyte entry is required for Alpha-synuclein induced inflammation and neurodegeneration in a model of Parkinson disease. Exp Neurol. 2018;300:179–87.
126. Stefanis L. α-Synuclein in Parkinson's disease. Cold Spring Harb Perspect Med. 2012;2(2):a009399.
127. Cook DA, Kannarkat GT, Cintron AF, Butkovich LM, Fraser KB, Chang J, et al. LRRK2 levels in immune cells are increased in Parkinson's disease. Npj Parkinson's Disease. 2017;3(1):11.
128. Atashrazm F, Hammond D, Perera G, Dobson-Stone C, Mueller N, Pickford R, et al. Reduced glucocerebrosidase activity in monocytes from patients with Parkinson's disease. Sci Rep. 2018;8(1):15446.
129. Perez-Pardo P, Dodiya HB, Engen PA, Forsyth CB, Huschens AM, Shaikh M, et al. Role of TLR4 in the gut-brain axis in Parkinson's disease: a translational study from men to mice. Gut. 2019;68(5):829–43.
130. Gerhard A, Pavese N, Hotton G, Turkheimer F, Es M, Hammers A, et al. In vivo imaging of microglial activation with [11C](R)-PK11195 PET in idiopathic Parkinson's disease. Neurobiol Dis. 2006;21(2):404–12.
131. Hirsch EC, Hunot S. Neuroinflammation in Parkinson's disease: a target for neuroprotection? Lancet Neurol. 2009;8(4):382–97.
132. Roussakis AA, Piccini P. Molecular imaging of Neuroinflammation in idiopathic Parkinson's disease. Int Rev Neurobiol. 2018;141:347–63.
133. Hui KY, Fernandez-Hernandez H, Hu J, Schaffner A, Pankratz N, Hsu N-Y, et al. Functional variants in the LRRK2 gene confer shared effects on risk for Crohn's disease and Parkinson's disease. Sci Transl Med. 2018;10(423):eaai7795.
134. Prigent A, Lionnet A, Durieu E, Chapelet G, Bourreille A, Neunlist M, et al. Enteric alpha-synuclein expression is increased in Crohn's disease. Acta Neuropathol. 2019;137(2):359–61.
135. Rivas MA, Avila BE, Koskela J, Huang H, Stevens C, Pirinen M, et al. Insights into the genetic epidemiology of Crohn's and rare diseases in the Ashkenazi Jewish population. PLoS Genet. 2018;14(5):e1007329.
136. Aliseychik MP, Andreeva TV, Rogaev EI. Immunogenetic factors of neurodegenerative diseases: the role of HLA class II. Biochemistry (Mosc). 2018;83(9):1104–16.
137. Hollenbach JA, Norman PJ, Creary LE, Damotte V, Montero-Martin G, Caillier S, et al. A specific amino acid motif of HLA-DRB1 mediates risk and interacts with smoking history in Parkinson's disease. Proc Natl Acad Sci U S A. 2019;116(15):7419–24.
138. Armstrong MJ, Okun MS. Diagnosis and treatment of Parkinson disease: a review. JAMA. 2020;323(6):548–60.
139. Agarwal S, Fleisher JE. Reaching those most in need—a call to action for advanced Parkinson's disease. Eur Neurol Rev. 2016;11(1):20–1.
140. Rajendran PR, Thompson RE, Reich SG. The use of alternative therapies by patients with Parkinson's disease. Neurology. 2001;57(5):790–4.

141. Shim J-M, Kim J. Cross-national differences in the holistic use of traditional east Asian medicine in East Asia. Health Promot Int. 2016;33(3):536–44.
142. Obeso JA, Stamelou M, Goetz CG, Poewe W, Lang AE, Weintraub D, et al. Past, present, and future of Parkinson's disease: a special essay on the 200th anniversary of the shaking palsy. Mov Disord. 2017;32(9):1264–310.
143. Curran J. The yellow Emperor's classic of internal medicine. BMJ. 2008;336(7647):777.
144. Li X, Zhang Y, Wang Y, Xu J, Xin P, Meng Y, et al. The mechanisms of traditional Chinese medicine underlying the prevention and treatment of Parkinson's disease. Front Pharmacol. 2017;8:634.
145. Chen H, Zhang Z, He J, Teng L, Yuan C. Traditional Chinese medicine symptom pattern analysis for Parkinson's disease. J Tradit Chin Med. 2017;37(5):688–94.
146. Sheng-d C. The protection of curcumin in nigral dopaminergic neuronal injury of mice model of Parkinson disease. Chin J Contemp Neurol Neurosurg. 2007;7(5):447–52.
147. Faust K, Gehrke S, Yang Y, Yang L, Beal MF, Lu B. Neuroprotective effects of compounds with antioxidant and anti-inflammatory properties in a drosophila model of Parkinson's disease. BMC Neurosci. 2009;10(1):109.
148. Cristian A, Katz M, Cutrone E, Walker RH. Evaluation of acupuncture in the treatment of Parkinson's disease: a double-blind pilot study. Mov Disord. 2005;20(9):1185–8.
149. Huang J, Qin X, Cai X, Huang Y. Effectiveness of acupuncture in the treatment of Parkinson's disease: an overview of systematic reviews. Front Neurol. 2020;11:917.
150. Wang H, Pan Y, Xue B, Wang X, Zhao F, Jia J, et al. The antioxidative effect of electro-acupuncture in a mouse model of Parkinson's disease. PLoS One. 2011;6(5):e19790.
151. Kang JM, Park HJ, Choi YG, Choe IH, Park JH, Kim YS, et al. Acupuncture inhibits microglial activation and inflammatory events in the MPTP-induced mouse model. Brain Res. 2007;1131(1):211–9.
152. Huo LR, Liang XB, Li B, Liang JT, He Y, Jia YJ, et al. The cortical and striatal gene expression profile of 100 hz electroacupuncture treatment in 6-hydroxydopamine-induced Parkinson's disease model. Evid Based Complement Alternat Med. 2012;2012:908439.
153. Chapelet G, Leclair-Visonneau L, Clairembault T, Neunlist M, Derkinderen P. Can the gut be the missing piece in uncovering PD pathogenesis? Parkinsonism Relat Disord. 2019;59:26–31.
154. Klingelhoefer L, Reichmann H. Pathogenesis of Parkinson disease—the gut-brain axis and environmental factors. Nat Rev Neurol. 2015;11(11):625–36.
155. Braak H, Rüb U, Gai WP, Del Tredici K. Idiopathic Parkinson's disease: possible routes by which vulnerable neuronal types may be subject to neuroinvasion by an unknown pathogen. J Neural Transm (Vienna). 2003;110(5):517–36.
156. Hawkes CH, Del Tredici K, Braak H. Parkinson's disease: a dual-hit hypothesis. Neuropathol Appl Neurobiol. 2007;33(6):599–614.
157. Kim S, Kwon SH, Kam TI, Panicker N, Karuppagounder SS, Lee S, et al. Transneuronal propagation of pathologic α-Synuclein from the gut to the brain models Parkinson's disease. Neuron. 2019;103(4):627–41.e7.
158. Svensson E, Horváth-Puhó E, Thomsen RW, Djurhuus JC, Pedersen L, Borghammer P, et al. Vagotomy and subsequent risk of Parkinson's disease. Ann Neurol. 2015;78(4):522–9.
159. Parashar A, Udayabanu M. Gut microbiota: implications in Parkinson's disease. Parkinsonism Relat Disord. 2017;38:1–7.
160. Rietdijk CD, Perez-Pardo P, Garssen J, van Wezel RJ, Kraneveld AD. Exploring Braak's hypothesis of Parkinson's disease. Front Neurol. 2017;8:37.
161. Su A, Gandhy R, Barlow C, Triadafilopoulos G. A practical review of gastrointestinal manifestations in Parkinson's disease. Parkinsonism Relat Disord. 2017;39:17–26.
162. Devos D, Lebouvier T, Lardeux B, Biraud M, Rouaud T, Pouclet H, et al. Colonic inflammation in Parkinson's disease. Neurobiol Dis. 2013;50:42–8.

163. Lin C-H, Chen C-C, Chiang H-L, Liou J-M, Chang C-M, Lu T-P, et al. Altered gut microbiota and inflammatory cytokine responses in patients with Parkinson's disease. J Neuroinflammation. 2019;16(1):129.
164. Jeon S, Kim YJ, Kim ST, Moon W, Chae Y, Kang M, et al. Proteomic analysis of the neuroprotective mechanisms of acupuncture treatment in a Parkinson's disease mouse model. Proteomics. 2008;8(22):4822–32.
165. Kim SN, Doo AR, Park JY, Bae H, Chae Y, Shim I, et al. Acupuncture enhances the synaptic dopamine availability to improve motor function in a mouse model of Parkinson's disease. PLoS One. 2011;6(11):e27566.
166. Kim SN, Kim ST, Doo AR, Park JY, Moon W, Chae Y, et al. Phosphatidylinositol 3-kinase/Akt signaling pathway mediates acupuncture-induced dopaminergic neuron protection and motor function improvement in a mouse model of Parkinson's disease. Int J Neurosci. 2011;121(10):562–9.
167. Park HJ, Lim S, Joo WS, Yin CS, Lee HS, Lee HJ, et al. Acupuncture prevents 6-hydroxydopamine-induced neuronal death in the nigrostriatal dopaminergic system in the rat Parkinson's disease model. Exp Neurol. 2003;180(1):93–8.
168. Park J-Y, Kim S-N, Yoo J, Jang J, Lee A, Oh J-Y, et al. Novel neuroprotective effects of melanin-concentrating hormone in Parkinson's disease. Mol Neurobiol. 2017;54(10):7706–21.
169. Park JY, Choi H, Baek S, Jang J, Lee A, Jeon S, et al. p53 signalling mediates acupuncture-induced neuroprotection in Parkinson's disease. Biochem Biophys Res Commun. 2015;460(3):772–9.
170. Tsai S-T, Wei T-H, Yang Y-W, Lu M-K, San S, Tsai C-H, et al. Transient receptor potential V1 modulates neuroinflammation in Parkinson's disease dementia: molecular implications for electroacupuncture and rivastigmine. Iran J Basic Med Sci. 2021;24(10):1336–45.
171. Torres-Rosas R, Yehia G, Peña G, Mishra P, del Rocio T-BM, Moreno-Eutimio MA, et al. Dopamine mediates vagal modulation of the immune system by electroacupuncture. Nat Med. 2014;20(3):291–5.
172. Jang J-H, Yeom M-J, Ahn S, Oh J-Y, Ji S, Kim T-H, et al. Acupuncture inhibits neuroinflammation and gut microbial dysbiosis in a mouse model of Parkinson's disease. Brain Behav Immun. 2020;89:641–55.
173. Kouti L, Noroozian M, Akhondzadeh S, Abdollahi M, Javadi MR, Faramarzi MA, et al. Nitric oxide and peroxynitrite serum levels in Parkinson's disease: correlation of oxidative stress and the severity of the disease. Eur Rev Med Pharmacol Sci. 2013;17(7):964–70.

# Anti-Inflammatory Therapy as a Promising Target in Neuropsychiatric Disorders

20

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**Abstract**

This chapter analyzes the therapeutic potential of current anti-inflammatory drugs in treating psychiatric diseases from a neuro-immunological perspective. Based on the bidirectional brain-immune system relationship, the rationale is that a dysregulated inflammation contributes to the pathogenesis of psychiatric and neurological disorders, while the immunology function is associated with psychological variables like stress, affective disorders, and psychosis. Under certain social, psychological, and environmental conditions and biological factors, a healthy inflammatory response and the associated "sickness behavior," which are aimed to resolve a physical injury and microbial threat, become harmful to the central nervous system. The features and mechanisms of the inflammatory response are described across the main mental illnesses with a special emphasis on the profile of cytokines and the function of the HPA axis. Next, it is reviewed the potential clinical utility of immunotherapy (cytokine agonists and antagonists), glucocorticoids, unconventional anti-inflammatory agents (statins, minocycline, statins, and polyunsaturated fatty acids (PUFAs)), the nonsteroidal anti-inflammatory drugs (NSAIDs), and particularly celecoxib, a selective cyclooxygenase-2 (Cox-2) inhibitor, as adjuvants of conventional psychiatric medications. The implementation of anti-inflammatory therapies holds great

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promise in psychiatry. Because the inflammatory background may account for the etiology and/or progression of psychiatric disorders only in a subset of patients, there is a need to elucidate the immune underpinnings of the mental illness progression, relapse, and remission. The identification of immune-related bio-signatures will ideally assist in the stratification of the psychiatric patient to predict the risk of mental disease, the prognosis, and the response to anti-inflammatory therapy.

**Keywords**

Anti-inflammatory agents · Cytokines · Glucocorticoids · Inflammation · Mental disorders · Psychoneuroimmunology

## 20.1 Introduction

Over 2000 years ago, Aristotle hypothesized a connection between physical health and mood. The concept reached its zenith with Descartes, who proposed the major advances in medicine, the Cartesian mind-body dualism. With the lighting up of psychoneuroimmunology in the 1980s of the past century, scientists began to explore one critical phenomenon of this dualistic paradigm, the interaction between psychological processes and the nervous and immune systems of the human body [1]. The immune system and the central nervous system maintain a relationship hitherto underestimated in many psychiatric illnesses. On one hand the immune system function is commonly associated with psychological variables like stress, distress, and affective disorders. Psychosocial stress together with cumulative genetic and epigenetic risk factors plays a role in the disturbances of the immune homeostasis [2]. On the other hand, a dysregulated inflammatory response of the immune system to harmful stimuli contributes to the pathogenesis of psychiatric and neurological disorders. This is the case of schizophrenia, autism spectrum disorders, bipolar disorders, depression, or even anorexia nervosa, whose neuropathological mechanisms may in some cases engage chronic inflammation [3–7]. Numerous epidemiological data have demonstrated the link that exists between a whole series of immune-inflammatory diseases and mental illnesses. For example, all the articles currently published on the microbiota and mental illnesses imply the same mechanism: inflammation increases the permeability of the digestive barrier and allows antigens to pass into the circulation which, normally, do not enter and will cause the appearance of autoantibodies and autoimmune diseases [8]. This is a phenomenon that is built up gradually, more or less quickly depending on the exposure. It is estimated that at least one third of patients with these severe conditions have elevated inflammatory markers. Some diseases heretofore considered to be exclusively psychiatric may also have a neurological and even immunological explanation. Understanding these psychiatric diseases from a neurological and immune perspective opens up new therapeutic possibilities [9].

Certain immunosuppressive treatments already known to treat multiple sclerosis or autoimmune encephalitis could find their place in the management of these mental conditions [10]. Trials currently being carried out with anti-inflammatory drugs associated with treatment, in particular in resistant depression [11], confirm our idea that this immuno-inflammatory pathway is extremely promising. This immunology approach is not only likely to move psychiatry towards neuropsychiatry but also to encourage healthcare professionals to look for signs of inflammation via certain additional examinations such as an MRI of the brain or assays in the blood and CSF of certain markers, such as C-reactive protein or CRP, and pro-inflammatory cytokines TNF-α or interleukin-6 (IL-6) [12, 13]. Many answers still remain to be found regarding the mechanisms of occurrence of these diseases and the effectiveness of anti-inflammatory treatments. To meet these challenges, the association of neurologists and psychiatrists in this new field, that is, immunoneuropsychiatry, seems promising for many patients. Recognition that inflammation may represent a common mechanism of disease extended to include neuropsychiatric disorders shakes up concepts in psychiatric illness.

An approach to exploring the connection between the immune system and mental condition is through medical illnesses associated with immune system dysfunction like HIV infection [14] and autoimmune disorders such as systemic lupus erythematosus (SLE) [15]. Immunomodulatory drugs have been known and have been used for many years to treat classic neurological autoimmune diseases such as multiple sclerosis and encephalitis. Pathogen-associated molecular patterns (PAMPs) and damage-associated molecular patterns (DAMPs) are induced by autoimmune disorders, toxins, infections, and even psychosocial stress to trigger both peripheral and central immune reactions through the binding to pattern recognition receptors (PRRs). The major consequences of ligating PRRs are to initiate a cascade of responses that direct inflammation, a process characterized by the activation of immune and non-immune cells that protect the host by eliminating threats and promoting tissue repair and recovery [16].

A healthy inflammatory response engages an acute immune response temporally restricted to the period the threat is present. Depending on the degree and extent of inflammation, specific energy-saving behaviors can occur that conserve metabolic energy and allocate more nutrients to the activated immune system. Biobehavioral effects of the immune system activation or “sickness behaviors” include sadness, anhedonia, fatigue, reduced libido and food intake, altered sleep, and social-behavioral withdrawal [17], which are critical for survival during times of physical injury and microbial threat. However, under certain social, psychological, environmental, and biological factors, the inflammatory response either fails to eliminate the damage or does not resolve once the threat has passed. Then, the process enters in a state of chronic, low-grade inflammation which is distinct from that in the onset. While acute inflammation is initiated by PAMPs, chronic inflammation is typically triggered by DAMPs in the absence of an acute physical insult and microbial threat. Shifts in the inflammatory response from short- to long-lived can cause a breakdown of immune tolerance [18].

## 20.2 Immune-Associated Pathophysiology of Mental Diseases

Inflammation aggresses the CNS and increases the risk for psychiatric disease [19]. Long considered to be protected by the immune system, the central nervous system (CNS), and in particular the brain, can also be the site of chronic inflammation. Cytokines are produced in CNS glial cells [20]. Astrocytes and microglia are key components of the innate immune system that can cause detrimental processes when activated while producing beneficial processes when quiescent. Cytokines increase neuronal excitotoxicity, reduce brain trophic factors and neurogenesis, and provoke oxidative stress directly by the release of reactive oxygen [3] and indirectly through the conversion of kynurenic acid, a product of the normal metabolism of amino acid L-tryptophan, to neurotoxic quinolinic acid (QA) and 3-hydroxykynurenine (3-HK) by activated microglia [21]. Although the entire region of brain parenchyma is excluded from the peripheral immune system, immune responses of the CNS are in close communication with the peripheral immune reactions [22, 23]. Circulating cytokines released by endothelial and immune cells in cerebral vasculature can diffuse passively or interact directly with BBB receptors stimulated by the central noradrenergic system to induce cyclooxygenase-2 (Cox-2) inflammatory signaling within the brain parenchyma. In addition, peripheral cytokines can also bind to receptors located on the liver, the spleen, or the nodose ganglion to relay cytokine signals to the brain via afferent sensory fibers of the vagus nerve to trigger neural firing or lead the synthesis of IL-6 by microglia [24, 25]. In the CNS, cytokines may also exert their effects by activating the hypothalamus-pituitary-adrenal (HPA) axis [26]. Given the abnormal profiles of pro-inflammatory and anti-inflammatory cytokines observed in some groups of psychiatric patients, an inappropriate CNS-immune communication may be the hallmark of neurodevelopmental, neurodegenerative, and neuro-immunomodulatory disorders.

How is immunity involved in the pathophysiology of mental diseases? Mental illnesses are due to the interaction between a genetic background and environmental factors. In the case of dysimmunity, the immunogenic background of the person does not allow him to defend himself sufficiently effectively against early environmental factors associated with the onset of mental pathologies, such as infections or severe stress, which are pro-inflammatory factors. This results in the appearance of an immuno-inflammatory cascade which varies according to the pathologies. Exposure to other environmental factors that are repeated throughout life, such as infections, stress, an unbalanced lifestyle (diet, physical activity, sleep, etc.), maintains this low-level inflammation, which will have consequences at the peripheral level, at the cerebral level, and on the digestive tract. Several lines of evidence suggest that dysfunction of innate immunity, including the microglia, the brain's resident immune cells derived from the monocyte lineage, may occur in a number of neuropsychiatric conditions [27]. Raised inflammatory processes (microglia activation and elevated cytokine levels) across diagnoses may disrupt neurobiological mechanisms regulating glutamate release and uptake, oxidative stress, and excitotoxicity [28]. Finally, cytokines activate the HPA axis to fuel inflammation and catecholaminergic neurotransmission [29].

### 20.2.1 Anxiety Disorders

Inflammation in the CNS primarily reflects physical and psychological stress. Early-life stress is more clearly associated with overt inflammation prior to the development of neuropsychiatric symptoms. For example, childhood trauma is associated with significantly elevated peripheral levels of C-reactive protein, IL-6, and TNF-α among other pro-inflammatory markers [30]. Stress can lead to increased cytokine levels and an induction of catecholamines via an activation of the HPA axis [29]. This in turn increases pro-inflammatory cytokines within and outside the CNS through a complex positive feedback loop [31]. Abnormalities in serotonergic function are involved in the pathogenesis of anxiety. The pro-inflammatory cytokines affect serotonin (5-HT) metabolism by reducing tryptophan levels. Cytokines appear to activate indoleamine-2-3-dioxygenase (IDO), an enzyme which metabolizes tryptophan, thereby reducing serotonin levels and creating neurotoxic serotoninergic metabolites 3-HK and QA, which next cause oxidative stress and permanent neuro-inflammatory damage [32]. Furthermore, inflammatory cytokines, such as IL-1β, may reduce extracellular 5-HT levels, via activation of 5-HT transporter mechanisms [33]. Disturbances in the microglial system increases TNF-α, oxygen radicals and oxidative stress [34], QA, and complement factors along with a decrease of neurotrophic factors of individuals genetically predisposed to hyper-anxiety [35].

Post-streptococcal autoimmune disorders are related to delayed neurological complications that persist throughout life in the function of the basal ganglia [5]. It would explain the enhanced pro-inflammatory innate immune response in the etiopathogenesis of obsessive compulsive disorders (OCD) [36]. The first evidence of the nexus between inflammation and OCD was found in the late 1980s, when the National Institute of Mental Health reported for the first time the association between streptococcal-induced Sydenham chorea and the abrupt, early-onset of obsessive-compulsive symptoms in pediatric patients. Although the syndrome was originally denominated pediatric autoimmune neuropsychiatric disorders associated with streptococcus or PANDAS [37], it has been reconsidered and evolved towards pediatric acute-onset neuropsychiatric syndrome (PANS) [38] and/or childhood acute neuropsychiatric syndrome (CANS) [39] all characterized by the presence of typical OCD symptoms and tics. In the case of adult OCD patients, it has been associated with a previous history of rheumatic fever following group A β-hemolytic streptococcal pharyngitis [40]. Other infectious agents like *Toxoplasma gondii* or Borna disease virus may also be of paramount importance to OCD. A complete picture of the changes in immune parameters in OCD is not possible owing to the scarce number of studies. Conversely to schizophrenia and BD, the circulation of the inflammatory cytokine IL-1β is decreased in patients with OCD [41]. Although this finding seems to indicate a non-inflammatory profile in OCD, this cytokine is likely to play a role in re-myelination, which is in agreement with the structural changes reported in OCD. The alterations of immune cells should be considered a state-dependent marker, perhaps related to stress associated with OCD. The

OCD-immune system relationship [42] hints for possible anti-inflammatory therapies in OCD [43].

Posttraumatic stress disorder or PTSD is a debilitating psychiatric disorder that follows trauma exposure. There is evidence that the immunological balance is skewed towards a pro-inflammatory state (IFN-γ, IL-6, TNF-α, and IL-17) in the plasma and increased levels of immune stimulatory Th1 and inflammatory Th17 cells in the blood following an initial trauma event [6]. Because of hyper-arousal state, people living with PTSD commonly manifest dysregulations of the systems that regulate the stress response, the HPA axis, and the sympatho-adrenomedullary system. The release of excess levels of stress hormones further contributes to low cortisol levels and chronic immune dysregulation in PTSD. This potentially causes the development of autoimmune disease, especially in younger individual. PTSD increase presents elevated risks for rheumatoid arthritis, systemic lupus erythematosus, multiple sclerosis, inflammatory bowel disease, and thyroiditis [44]. Interestingly, among the genetic variations associated with the risk for PTSD after trauma exposure, there are included genes encoding regulators of the immune function [45]. Enhanced cell-mediated immune function and pro-inflammatory cytokine level significantly increase the odds of developing clinically worse courses of PTSD. For instance, there is a direct correlation between PTSD severity and spontaneous overnight secretion of IL-6 and TNF-α by the leukocytes [46]. Because of the close association between PTSD and neuroendocrine and immune dysfunction, and given the increased risk for comorbid somatic autoimmune and inflammatory disorders in PTSD, the targeting of the neuroendocrine and immune dysfunction is likely to improve PTSD symptoms [47].

### 20.2.2 Mood-Related Disorders

Altered cytokine activity in the periphery and the brain of a subpopulation of depressed patients [48] brings support to the concept of depression-associated inflammation [49]. Patients with major depressive disorders (MDD) demonstrate that C-reactive protein and inflammatory cytokines are strongly correlated with CSF markers of neuro-inflammation, which suggests that peripheral inflammatory biomarkers may reflect similar findings in the CNS [50]. Psychosocial stress, a well-known precipitant of mood disorders, is capable of stimulating neuro-inflammatory pathways within the brain [3], while MDD occurs at a substantially higher rate in patients with inflammatory disorders in peripheral organs such as multiple sclerosis, psoriasis, rheumatoid arthritis, inflammatory bowel disease, and myocardial infarction. Individuals with autoimmune diseases who are given inflammation-based therapies (e.g., interferon, typhoid vaccination, or endotoxin) are at an increased risk of presenting with mood disorders [51]. When used for immunotherapy in cancer or hepatitis, large doses of pro-inflammatory IL-2 and/or IFN-α induce depressive symptoms that can be efficiently treated by antidepressants [52, 53]. The overactivation of the immune system over the course of life (e.g., aging-related and comorbid disease-related inflammatory processes) also increases

the vulnerability to anxiety and depression. In accordance with the phenotypic heterogeneity of MDD, a pattern of low-grade inflammation is present in at least one third of MDD cases, with being atypical depression a more pro-inflammatory condition [54]. Somatic or neuro-vegetative symptoms of depression (fatigue, sleep disturbances, poor appetite) are more associated with inflammation than emotional/cognitive symptoms (depressed mood, worthlessness, anhedonia, poor concentration). In this vein, depression probably represents a maladaptive version of "sickness behavior" (social withdrawal, reduced appetite, and low energy), which might occur in the presence of an exacerbation in intensity and/or duration of the innate immune response [55].

Small physiologic differences in the immune system can have a huge effect over time on depression if they are consistently skewed in one direction. It has been hypothesized that the activation of microglia from stress or preexisting pro-inflammatory state causes metabolic changes in the tryptophan-kynurenine pathway [56]. Tryptophan is the main precursor of 5-HT, whose deficiency leads to depression, whereas kynurenine is the precursor of the neuroprotective molecule kynurenic acid that antagonizes the NMDA receptor. However, pro-inflammatory cytokines activate the IDO enzyme, which metabolizes kynurenine into excitotoxic metabolites like 3-HK and QA [21]. Oxidative stress induced by the overweight of N-methyl-D-aspartate (NMDA) agonism leads to the loss of glial elements, altered glutamate release/reuptake, and decreased neurotrophic support that characterize depressive disorders. Cytokines cause tryptophan depletion by the stimulation of the IDO synthesis and the promotion of the neurotoxic pathway of the kynurenine pathway [32]. Another pivotal mechanism by which cytokines may induce depression is the activation of the HPA axis [57]. Pro-inflammatory cytokines like IL-1, IL-6, TNF-α, and IFN-γ can result in the synthesis of corticotrophin-releasing factor (CRF), which in turn stimulates adrenocorticotropic hormone (ACTH) release and the subsequent hyperactivity of the HPA axis [26]. Clinical studies have demonstrated hyperactivity of the HPA axis and increased levels of cortisol in patients with major depression, because of an impairment of glucocorticoid receptor (GR)-mediated negative feedback or glucocorticoid resistance [58]. Reduction of GR function is the main neuroendocrine abnormality in depression, and hypercortisolemia is seen as a compensatory mechanism in the presence of reduced brain sensitivity to glucocorticoids. Although corticosteroids are generally anti-inflammatory, at normal endogenous levels, adrenal steroids appear to function as immune regulators rather than simply immune suppressors [59]. A lack of the "positive" effects of cortisol on the brain, because of "glucocorticoid resistance," is likely to be involved in the pathogenesis of melancholic depression.

Chronic (low-grade) dysregulated immune activation (e.g., Guillain-Barré syndrome, autoimmune hepatitis, multiple sclerosis, rheumatoid arthritis, systemic lupus erythematosus, psoriasis, and autoimmune thyroiditis) in patients suffering from bipolar disorder (BD) is present at higher rates than in normal population, suggesting a significant cross-talk between autoimmune processes and BD [60]. Indeed, patients with BD exhibit increased rates of obesity and metabolic syndrome, conditions associated with low-grade inflammation. Immune-BD

interaction may be bidirectional. BD increases the risk of the development of comorbidities, such as cardiovascular and metabolic diseases. Increased and decreased levels of IL-1β and IL-6 respectively in the cerebrospinal fluid of a subset of BD patients during mania and depression are suggestive of the CNS-focused immune mechanisms [61]. BD patients present a hypo-responsive glucocorticoid receptor (GR) in peripheral tissues, which could be at least partly responsible for a deficient cortisol-mediated negative feedback loop of the HPA axis and basal hypercortisolemia. Pro-inflammatory cytokines contribute to a chronic HPA activation and inflammatory responses in BD [62].

Finally, immune deficiencies are secondary processes to malnutrition observed in the development and progression of another mood-related disorder, anorexia nervosa [63]. During the course of anorexia, there are metabolic changes; hormonal imbalances, particularly with regard to secretion of cortisol; and altered production of various neurotransmitters, which result in a dysfunctional immune system [64]. Malnutrition causes a significant reduction in the percentage of T-cells and unchanged or slightly elevated B-cell numbers which provokes a high instability in the immune system of anorexia patients [65]. Consequently, the severity of anorexia nervosa correlates with higher levels of peripheral inflammatory markers [66]. Plasma levels of the pro-inflammatory cytokines TNF-α and IL-1β in plasma significantly increase in patients with anorexia nervosa, while the levels of prostaglandins $PGE_2$ (pro-inflammatory) and 15d-$PGJ_2$, (anti-inflammatory) and their PPARγ receptor implicated in their modulation diminish [7]. Cytokines directly interact with hunger centers, especially IL-1 and TNF-α, which additionally affects peripheral signals to satiety centers, leading to temporary gastric emptying inhibition. Of especial interest are the adverse effects of increased TNF production on anorexia, since this cytokine has an exacerbating central effect of food intake suppression and/or increased tissue catabolism [67]. Despite the immune deficiencies and changes in immunological parameters, an unexplained, remaining paradox is that many people suffering from anorexia appear very healthy and do not suffer from viral infections excluding cases of advanced malnutrition.

### 20.2.3 Schizophrenia

Schizophrenia is a neurodevelopmental disease driven by risk genes, and the immune system is positioned as a common link between the seemingly diverse genetic and environmental risk factors for schizophrenia [68]. Appearance of psychotic symptoms represents a relatively late manifestation triggered by environmental stress factors and disturbances of the immune system. Accordingly, the contribution of the immune dysregulation to the pathogenesis of schizophrenia may occur even before the onset of full-blown psychosis [69]. Schizophrenic patients with a history of prenatal exposure to influenza infection (second trimester) and rubella infection (first trimester) show impaired neurocognitive performance and structural abnormalities (e.g., synaptic pruning) in the brain [70]. The critical mediators of neuro-inflammation IL-6, which is highly expressed in fetal brains

following maternal immune activation [71], and IL-1β alter the neuronal development of the dopaminergic and serotonergic systems [72], thus causing functional deficits in the brain. These alterations may even prime the innate CNS immunocompetent cells so that they would later on exaggerate inflammatory responses. This would explain why physical and mental stress, HIV and influenza infections, and autoimmune disorders such as systemic lupus erythematosus (SLE) are associated with psychotic symptoms in more vulnerable individuals. Pro-inflammatory cytokines like IL-1β, IL-6, and TNF-α are increased in the peripheral blood of patients with schizophrenia during acute psychotic exacerbations and related to a greater severity of both cognitive deficits and negative symptoms [73]. TNF-α and IL-6 cross the blood-brain barrier (BBB) and modulate several molecular/cellular processes, including, but not limited to, monoamine metabolism. It suggests that immunological alterations may even affect their clinical status after the onset of the illness.

There is an intertwined interaction of pro-inflammatory cytokines with the dopaminergic and glutamatergic neurotransmitter systems in areas affected by schizophrenia like the prefrontal cortex and hippocampus [74]. Conversely to depression, type-1 immune responses (e.g., IL-2 release) is blunted in schizophrenia, which may lead to an unbalance in IDO and in the tryptophan-kynurenine metabolism associated with an imbalance in the glutamatergic neurotransmission and NMDA antagonism in schizophrenia [75]. In addition, low concentrations of IL-2 may also alter dopamine-mediated neurotransmission. Neuroleptic medications used for psychosis also influence immune factors, often normalizing or reversing the direction of the abnormalities described in premedicated patients. Neuroleptic administration is associated with type-1 activation, including decreased IL-6 and soluble IL-6 receptors (sIL-6R), normalization of IFN-γ production, and increased sIL-2R. Nevertheless, recent studies suggest that the cytokine profile changes with the clinical status of the patients, with a high level of pro-inflammatory cytokines like IL-1β, IL-6, and transforming growth factor-beta (TGF-β) during the acute phase of diseases, which is absent in the remission phase [76].

### 20.2.4 Autism Disorders

In the case of autism, pro-inflammatory factors, infections, or autoimmune diseases are most likely involved during pregnancy, leading to genetically predisposed fetuses to develop this condition before the age of three [77]. The first evidence of the familial link of polyendocrine autoimmune disorder with autism was reported 50 years ago. Since then, some large population-based studies support the theory that autoimmune responses and immune dysfunction at or around the time of pregnancy may be related to a later diagnosis of autism in the offspring [78]. For instance, increased rates of rheumatoid arthritis, celiac disease, psoriasis, and type 1 diabetes, as well as immune-mediated disorders such as asthma and allergies, are found in mothers of children with autism. In addition, animal models known as immune activation in the mother, in which inflammation is induced through infections during

pregnancy, trigger the appearance of autism-mimicking behaviors in offspring. Accordingly, global immune dysfunction in mothers during pregnancy, rather than specific diseases, may be associated with increased risk for autism disorder. This risk nexus is not limited to the mothers, since a higher rate of the autoimmune condition type 1 diabetes is reported in fathers, which suggests underlying heritable immunogenetic factors [79]. Nonetheless, observations of autoimmunity are not limited to families of the children with autism disorder, but also to the presence of immune dysfunction in some children with autism disorder. Increased levels of pro-inflammatory cytokines such as IL-6 and TNF-α in brain specimens and CSF, as well as in the periphery in autism disorder individuals, point that to an ongoing neuroinflammatory process in autism disorder [80]. The presence of antibodies directed against adult brain or CNS tissue, but not fetal brain tissue, has been repeatedly reported in children with autism disorder [77]. Findings so far published suggest a complex pattern of immune activation that varies among different subgroups of individuals with autism disorder. Unfortunately, the extent of immune abnormalities in the broader autism disorder phenotype is not yet well understood, neither are known the mechanisms by which immune dysfunction contributes to the etiology of autism disorder.

## 20.3 Anti-Inflammatory and Immune-Based Therapies for Treatment-Resistant Mental Illness

Psychiatric disorders present a tremendously large heterogeneity that accounts for the lack of responsiveness and high rates of treatment resistance to conventional neuroleptic, antidepressant, and anxiolytic drugs. Although the monoaminergic hypothesis has been dominant in our understanding of the pharmacological effects of psychotropic medications, additional mechanisms might also play a role. Neurotransmitters involved in the neurobiology of mental health and disease like dopamine, serotonin, and glutamate have been found altered in low-level neuroinflammation. Therefore, dysfunction of the immune system and brain-immune interactions may be some of the sources of the neurotransmitter deficits historically ascribed to the major mental disorders [81]. In recent years, there has been a paradigm shift to place abnormal cytokine profiles at the center of psychiatric symptoms. Pro-inflammatory cytokine levels like TNF-α and IL-6 are related to the level of mental distress in some psychiatric inpatients suggesting that low-grade inflammation is probably a cause of resistance to conventional pharmacological treatments [54, 82]. In this vein, some recent studies have shown promising results with anti-inflammatory therapies like steroids, plasmapheresis, intravenous immunoglobulin, cyclophosphamide, or monoclonal antibodies acting on B cells, particularly in the treatment of certain children with autism, who suffer from inflammation, or adults with schizophrenia, for whom immunosuppressive therapy or a bone marrow transplant has significantly reduced psychiatric symptoms [83]. Some studies have even made it possible to highlight the anti-inflammatory role, hitherto unknown, of successful antidepressant treatment like selective serotonin reuptake

inhibitors widely used today [84]. The antidepressant bupropion interferes with the production of cytokines, while antipsychotic drugs like clozapine, risperidone, and haloperidol influence the balance between anti-inflammatory and pro-inflammatory cytokines upon stimulation of the immune system. In the light of this, drugs with demonstrated anti-inflammatory effects may well show improvement of mental conditions when used as add-on treatments to conventional psychiatric medications [85–91]. Increasing evidence demonstrates that anti-inflammatory agents are likely to modify the relationship between cytokines and mental distress (Table 20.1). Nonetheless, no superiority has been found in anti-inflammatory monotherapy, raising the question of the mechanism behind the effect.

### 20.3.1 Cytokine Antagonists and Agonists

Given their specificity, immunotherapy against cytokines offers an unparalleled opportunity to directly test the hypothesis of whether immune dysfunctions play a causal role in psychopathology. The use in schizophrenia of monoclonal antibodies like natalizumab, siltuximab¸ canakinumab, and tocilizumab targeting specific immune molecules is an illustrative example [92]. The same holds true for the treatment of depression. Anti-TNF therapy, which is being considered as an option in improving postoperative cognitive dysfunction, has shown clinical efficacy on cognition and depressive symptoms [93]. However, the complex signaling pathways of TNF-α and its receptors and the duality of its function in being both neuroprotective and neurodegenerative preclude long-term benefits of anti-TNF-α therapies [94]. The pro-inflammatory cytokine IFN-γ plays a pivotal role in modulating immune and inflammatory responses. The effect of IFN-γ-1b on stimulating the type-1 immune response showed preliminary, but encouraging, results in reducing clinical symptoms of schizophrenia [95]. Immunotherapy may also have possible psychiatric adverse effects: there is evidence that the treatment of hepatitis C with IFN-α precipitates depressive episodes [96]. Before considering immunotherapy as an adjunctive to conventional psychotropic medications, there is the need to improve our understanding of cytokine actions in the CNS and how peripheral inflammation reflects or perpetuates psychiatric symptoms.

### 20.3.2 Glucocorticoids

Glucocorticoids produced by the zona fasciculata of the adrenal cortex are a class of steroid hormones that are part of the feedback mechanisms of the immune system. Glucocorticoids are often exploited for their immune-suppressor properties [97], since they inhibit prostaglandins (PGs) and leukotrienes, the two main products of inflammation. Glucocorticoids act at the level of phospholipase $A_2$ ($PLA_2$), the enzyme that supplies the arachidonic acid (AA) substrate to both cyclooxygenase/PGE isomerase (COX-1 and COX-2 isoenzymes), to synthetize prostaglandin $H_2$ ($PGH_2$), and to the lipoxygenases that catalyze the dioxygenation of AA in a class of

**Table 20.1** Evidence for anti-inflammatory therapies in neuropsychiatric disorders

| Mental disorder | Immune-associated pathophysiology | Anti-inflammatory drug [Ref.no.] |
|---|---|---|
| Anxiety | Catecholamine depletion and excessive oxidative stress due to elevated cytokines via HPA axis | NAC [109, 111]<br>Aspirin [161]<br>Diclofenac [162]<br>Naproxen [162]<br>Ketoprofen [162] |
| OCD | Autoimmune syndrome largely caused by β-hemolytic streptococcal infections | NAC [109]<br>Celecoxib [154] |
| PTSD | Autoimmune dysregulation caused by excessive levels of stress hormones | Glucocorticoids [106, 107] |
| MDD | Low-grade chronic inflammation caused by aging-related and comorbid disease-related inflammatory processes | Anti-TNF-α [93]<br>Minocycline [115]<br>Omega-3 FA [125, 126]<br>Probiotics [128]<br>Celecoxib [150–153]<br>NSAIDs [161, 162] |
| BD | Autoimmune diseases and low-grade chronic inflammation | Minocycline [116]<br>Celecoxib [116, 155] |
| Anorexia nervosa | Dysfunctional immune system caused by malnutrition | Unknown |
| Schizophrenia and psychosis | Immune dysregulation caused by perinatal influenza and HIV infections and autoimmune disorders | Cytokine monoclonal Ab [92]<br>NAC [110, 158]<br>Statins [122]<br>Omega-3 FA [124]<br>Celecoxib [138, 151, 159]<br>Minocycline [158]<br>Aspirin [158, 160] |
| Autism-related disorders | Infections and autoimmune disease during pregnancy | Celecoxib [157] |

*Ab* antibody, *BD* bipolar disorder, *FA* fatty acids, *MDD* major depressive disorder, *NAC* N-acetylcysteine, *NSAIDs* nonsteroidal anti-inflammatory drugs, *OCD* obsessive compulsive disorder, *PTSD* posttraumatic stress disorder

lipids called leukotrienes characterized by containing a cis, cis-1,4-pentadiene. In addition, glucocorticoids also inhibit both COX isoenzymes, an effect being much like that of NSAIDs (see next section). Finally, glucocorticoids suppress COX expression, which reinforces their anti-inflammatory effects.

How endogenous or exogenous glucocorticoids, through their immune and inflammatory inhibiting or promoting properties, would alter brain function and

behavior is unknown and requires investigation. At normal endogenous levels, adrenal steroids appear to function as immune modulators [98]. They shift cytokine production to favor the type-2 immune response while inhibiting type-1 response. Chronic or acute stress and Cushing's disease can produce an excess of endogenous corticosteroids, thus increasing susceptibility to mood changes, cognitive deficits, and even psychosis [99]. Likewise, acute corticosteroid treatment with prednisone and dexamethasone adversely impacts memory, executive functions, and mood [100]. Exacerbated glucocorticoid levels cause neuronal damage and lasting alterations to the plasticity and structural integrity of the hippocampus and prefrontal cortex, and this mechanism may plausibly contribute to impaired memory and cognition in critical illness survivors [101] and in children and adolescents with inflammatory bowel disease [100]. Among the behavioral outcomes of high glucocorticoids, mood and anhedonia appeared to be the most consistently and strongly affected [102]. Chronic stress primes neuro-inflammatory responses in a glucocorticoid-dependent manner [103]. Therefore, the glucocorticoid state of the patient preceding illness may be important for the eventual outcome. According to the glucocorticoid resistance hypothesis of depression [104], increased levels of cortisol may be the consequence of an impairment of glucocorticoid receptor (GR)-mediated negative feedback on the HPA axis. Rather than using immunosuppressive corticoid-based treatment, the therapy of depression and mood-related disorders may well benefit from manipulating GR function with both agonists and antagonists. Conversely, glucocorticoid-based therapy can possibly protect against the development of PTSD given the association with low cortisol levels. Glucocorticoid treatment at the time of acute stress may prevent changes in hippocampal and amygdala architecture and associated changes in affective behavior [105]. The exogenous treatment with glucocorticoids has shown promise for the prevention of PTSD after a traumatic experience [106]. High doses of glucocorticoids administered with appropriate timing may block fear memory formation or retrieval, although moderate doses would also be expected to enhance fear memory, depending on their timing [107].

### 20.3.3 Unconventional Anti-Inflammatory Agents

N-Acetylcysteine (NAC) is a synthetic derivative of the endogenous amino acid L-cysteine and a precursor of glutathione with well-known anti-inflammatory and antioxidant properties [108]. Several studies have demonstrated that NAC regulates impaired glutamate and dopamine neurotransmission. There is preliminary, but encouraging, evidence of the therapeutic potential of NAC in disorders such as anxiety and attention deficit hyperactivity disorder [109]. Some evidence exists to support the use of NAC as an adjunct treatment to reduce the total and negative symptoms of schizophrenia [110]. In addition, NAC also appears to be effective in reducing craving in substance use disorders, especially cocaine and cannabis [111].

Minocycline is a tetracycline antibiotic with potential as an adjunctive treatment in psychiatry [112] due to its anti-inflammatory and anti-apoptotic/neuroprotective

properties and inhibition of cytochrome P450 enzymes that metabolize antipsychotics such as clozapine [113]. Minocycline has been checked in open-label or small randomized controlled trials in psychiatry showing divergent outcomes, with positive results in some studies counterbalanced by a number of cases with no significant improvements [114]. Anecdotal evidence supports minocycline's efficacy for augmentation of antidepressants in treatment-resistant depression patients with low-grade peripheral inflammation [115]. Minocycline may potentially be useful as an adjunctive for BD [116]. There is no evidence that minocycline or celecoxib monotherapy was superior to placebo for the treatment of BD. Minocycline reduces fear processing and improves implicit learning in healthy volunteers [117] and may still hold promise like a candidate treatment for depression owing to its neuroprotective role.

Statins are cholesterol-lowering agents that act by inhibiting 3-hydroxy-3-methylglutaryl coenzyme A reductase. Several studies have suggested that statins may have anti-inflammatory properties, with lowering pro-inflammatory markers such as IL-1β, IL-6, TNF-α, and C-reactive protein levels [118]. Simvastatin also could alleviate cognitive function, since it modulates muscarinic $M_1$ and $M_4$ receptors, central dopamine $D_1$ and $D_2$ receptors, and the serotonin transporter [119]. Whereas conflicting evidence exists about the relationship between statins and mood amelioration [120, 121], a meta-analysis of statin adjunctive therapy for schizophrenia showed that statins improved the Positive and Negative Syndrome Scale (PANSS) [122].

Polyunsaturated fatty acids like omega-3 present antioxidation, anti-inflammation, and neuroprotection. In humans, dietary deficiencies of omega-3 fatty acids, in particular eicosapentaenoic and docosahexaenoic acids, have been linked to increased risk of developing MDD, BD, schizophrenia, dementia, attention deficit hyperactivity disorder, and autism [123]. Diet omega-3 fatty acids are essential because of their anti-inflammatory, antioxidative, and neuroprotective effects on neuronal membrane fluidity. Randomized clinical trials have found a significant benefit of omega-3 adjunctive schizophrenia therapy in the total, positive, and negative PANSS scores of patients or in their cognitive function [124]. For the remaining psychiatric disturbances, the data are too scarce to draw any conclusion regarding the benefits of diet supplementation with omega-3 fatty acids. Omega-3 fatty acid replacement therapy has only been shown to have a mild effect for the treatment of mood disorders and ADHD [125, 126].

Probiotics have traditionally been used to reestablish the physiological functions of the gastrointestinal tract. Given the extensive bidirectional communication between the gastrointestinal tract and the CNS, the gut-brain axis [127], probiotics are capable of changing the behavior and decreasing the levels of systemic inflammatory markers in animal models. A meta-analysis of randomized controlled trials has suggested that probiotics may be associated with a significant reduction in depression [128].

## 20.4 Nonsteroidal Anti-Inflammatory Drugs (NSAIDs): The Targeting of Cox-2

Nonsteroidal anti-inflammatory drugs (NSAIDs) are used as antipyretic, anti-inflammatory, and analgesic agents. NSAIDs work by inhibiting the activity of cyclooxygenase Cox-1 or Cox-2 isoenzymes [129]. There are two types of NSAIDs available: nonselective and Cox-2-selective inhibitors (celecoxib, etoricoxib). Non-selective NSAIDs are typically divided into groups based on their chemical structure: acetylated salicylates (aspirin), non-acetylated salicylates (diflunisal, salsalate), propionic acids (naproxen, ibuprofen), acetic acids (diclofenac, indomethacin), enolic acids (meloxicam, piroxicam), anthranilic acids (meclofenamate, mefenamic acid), and naphthylalanine (nabumetone). Cyclooxygenases (Cox) are a group of heme-containing enzymes that catalyze a rate-limiting conversion of AA to largely bioactive prostaglandins (PGs) involved in inflammation, through the addition of molecular oxygen [130]. The Cox-1 isoform is a housekeeping enzyme constitutively expressed in all tissues. Although the mitogen-inducible Cox-2 is activated to cause inflammation, this isoform is also constitutively expressed in certain tissues as in the kidney and in the brain. In the human brain, Cox-1 is preferentially expressed in microglia, where Cox-2 is in glutamatergic neurons in the cerebral cortex, hippocampus, and amygdala [131]. Prostaglandin $PGE_2$ may be the predominant metabolite of the enzymatic activity of Cox-2 in the brain where it may function as neuromodulators of the inflammation as well as may be involved in important physiology functions in synaptic plasticity and long-term potentiation [132]. Immunological disturbances may lead to an increased $PGE_2$ production and probably also in an increased Cox-2 expression that would also contribute to neuropathology by enhancing glutamate excitotoxicity [133], promoting neuronal cell death, and metabolizing the endogenous cannabinoid 2-arachidonoyl-glycerol into $PGE_{2G}$ through the oxidation of its AA moiety. Some studies suggest upregulation of Cox-2 expression in inflammatory and neurodegenerative diseases [134] as well as schizophrenia [135] and bipolar disorder [136].

NSAIDs penetrate the brain [137], and the use of NSAIDs as adjunctive treatments in neuropsychiatric disorders—including schizophrenia, bipolar disorder, and major depressive disorder—is currently under investigation [138–141]. NSAID treatment benefits on the brain in depression are thought to be due to their ability to block Cox-1 during pro-inflammatory microglial activation and neuronal Cox-2, which may affect glutamatergic and monoaminergic neurotransmission [133]. Cox-2 expression is upregulated in inflammatory schizophrenia and bipolar disorder. Accordingly, the selective Cox-2 inhibitor celecoxib has so far been the most studied NSAIDs in psychiatry [142, 143] because of the inhibition of microglial activation and glutamate release, the enhancement of serotonergic and noradrenergic output in the prefrontal cortex, and the modulation of glucocorticoid receptors (off-target mechanism of action). When reviewed the literature to determine whether selective Cox-2 and nonselective Cox inhibitor NSAIDs as adjuncts or monotherapy affect depressive symptoms [144–146], the search gives mix results regarding efficacy. Possible confounding factors [147] include age range (young versus elderly

subjects), sex, presence of antidepressant use, medical comorbidities (diabetes, metabolic syndrome), method of depression measure (somatic symptoms are more sensitive than subjective feelings to the influence of NSAIDs), severity of depressive symptoms, clinical phase of the illness (most of the studies rest on trials in acute depression), duration and study design (randomized controlled trials, cohort studies, and an open label), and pharmacological strategies (add-on treatments versus monotherapy). Despite the negligible therapeutic effects of NSAIDs reported by one meta-analysis in MDD [148], celecoxib reaches the CNS in humans in concentrations sufficient to inhibit Cox-2 [149] and thus improve the therapeutic management of depression [150–153], BD [117], and schizophrenia [138]. In effect, celecoxib works as an adjunctive treatment to fluvoxamine in moderate to severe OCD [154], to escitalopram in treatment-resistant BD [155], and to reboxetine and vortioxetine in MDD [156]. The combination of risperidone and celecoxib is superior to risperidone alone in treating irritability, social withdrawal, and stereotypy of children with autism [157]. In schizophrenia, celecoxib has shown efficacy in augmentation of amisulpride treatment in the early disease stages and first psychosis episode [158] as well as an effective adjuvant agent to risperidone in the management of patients with chronic schizophrenia [159]. It should be noticed that the use celecoxib in the treatment of schizophrenia reduces the symptoms only when administered in combination with the anti-schizophrenic drugs (i.e., risperidone, olanzapine, amisulpride). In the fact of BD, celecoxib monotherapy is not superior to placebo either. Strikingly, aspirin, which is a nonselective Cox inhibitor with preferential selectivity for the Cox-1 isoenzyme, significantly reduced the positive and negative symptoms of schizophrenia regardless it is administered either alone or as adjunctive therapy [160]. In a large register-based cohort study in Sweden, aspirin and other NSAIDs have demonstrated their effectivity in decreasing the risk of depression, anxiety, and stress-related disorders during the first year following cancer diagnosis [161]. A population-scale retrospective analysis has demonstrated the anxiolytic effects of ketoprofen, diclofenac, and naproxen in patients with pain [162].

Despite these promising preliminary results, the efficacy and safety of chronic NSAID exposure have been called into question in the treatment of both symptoms of depression [163], particularly in the elderly [164], and of psychotic disorders [148, 165]. The conflicting evidence may be due to the methodological heterogeneity of the clinical trials and the selection bias (inadequate assessment of the inflammatory and clinical status of patients). Another neglected aspect is that Cox selectivity of NSAIDs matters. Although neuro-inflammation is originally triggered by the induction of glial Cox-2 expression, the activity of Cox-1 also yields a prooxidant/pro-inflammatory action. Neuronal Cox-2 plays a homeostatic role in synaptic transmission and plasticity [133]. Deviations in inflammatory levels in both directions may actually impair neural plasticity. Studies show that both inflammation and neural plasticity act as key players in the vulnerability and recovery from psychiatric disorders with an impact on anxiety and memory [166]. Accordingly, a failure in the Cox-2/Cox-1 ratio might cause behavioral disturbances that otherwise would be commonly ascribed to neuro-inflammation [167].

Blanket blockade of Cox-2 may not be advisable because Cox-2 expression might in fact have pro-resolution properties [168]. In addition, selective Cox-2 inhibitors may alter the metabolism of the endocannabinoid system of the brain [169]. Given the significance of different Cox isoforms and their unknown role of their relative levels in the CNS, careful attention must be given to selection and evaluation of specific NSAIDs. Aspirin whose activity on Cox-1 prevails over Cox-2 alleviates psychiatric symptoms on its own (Hu et al., 2020). An interesting alternative to Cox inhibition would be the pharmacological intervention of the AA cascade. Some genetic evidence supports the notion that disturbances of the $PLA_2$-Cox-2 axis underlie abnormalities of monoaminergic neurotransmission in schizophrenia [170], BD [171, 172], and MDD [173]. In addition, preclinical experiments have also confirmed that mood stabilizers like lithium chloride target the upstream release of AA substrate for Cox enzymes [174].

## 20.5 Final Remarks

Although we now know by decades of research that there is a robust and complex link between inflammation and mental illness, one must be cautious about the apparent simplicity of the idea that anti-inflammatory agents could improve psychiatric symptoms. Meta-analyses do not undermine the potential clinical utility of anti-inflammatory agents, but they suggest that clinical trials carry a variety of caveats that need careful consideration. What the reviewed cohort studies and follow-up studies have actually demonstrated is that the inflammation-mental health link lacks diagnostic specificity and varies considerably among individuals and with each clinical phase of illness. Given the multifactorial etiology, preexisting inflammatory conditions may then account for at least a subset of psychiatric patients. Moreover, the biology of inflammation and related immune alteration may depend on the stage of the illness as it does the clinical symptomatology. For example, in schizophrenia and BD, a marked inflammation appears during episodes of acute decompensation so that chronic, low-grade inflammation seems to precede the initial illness episode [69]. Most of the studies in the field have not considered the inflammatory status before starting the anti-inflammatory clinical trials. Finally, while the onset of mental disorders appears well explained by its inflammatory background, the immune underpinnings of their progression, relapse, and remission remain to be elucidated. In some cases, anti-inflammatory treatments used outside the acute clinical phase may be detrimental because of the ambivalence nature of the inflammatory response.

There are some interesting future prospects to undertake the difficulties found in implementing anti-inflammatory therapies in psychiatry. Firstly, a number of publications indicate the importance of stratifying patients on the basis of their degree of phase-specific neuro-immune dysfunction and surrogate biological signatures of inflammation [12, 175–177] aided by neuroimaging to launch therapeutic trials. The identification of immune-related bio-signatures will ideally assist in predicting risk of disease, prognosis, and response to therapy. A broad immune-phenotyping is likely to be essential to identify the subpopulations of psychiatric

patients who are likely to respond to anti-inflammatory therapy either alone or when combined with conventional psychiatric drugs. In the second place, there are important gaps in our knowledge about the immune-associated pathophysiology. For example, the majority of studies investigating the role of inflammation in psychiatry conditions assessed peripheral levels (i.e., plasma or serum) of cytokines, while only a few studies evaluated CSF cytokine levels [178, 179], which may reflect better CNS levels and, therefore, any ongoing neuro-inflammatory process. Surprisingly, microglia activation shows no significant association with specific diagnostic categories of mental conditions [180], which means that it is not present in all psychiatric patients. It should then be explored how peripheral and neural immune mechanisms interact in these cases, particularly at the level of the blood-brain barrier as well the dynamics of the innate and adaptive immune responses. Finally, in the advent of precision medicine in psychiatry, it is important to understand the pharmacological off-target effects of the anti-inflammatory agents described in this chapter. The enhanced neuroprotection plus a reduction in inflammation may be an extended avenue for future interventions at least in depression. The complex opposing functions of TNF-α (neuroprotective and neurodegenerative) advice against the long-term benefits of anti-TNF-α therapies [181]. The lack of knowledge on immune-physiology and neurobiology of Cox enzymes limits the therapeutic potential of selective Cox-2 inhibitors, since Cox-1 is also pro-inflammatory.

In summary, anti-inflammatory pharmacotherapy may need to be used according to the phase of illness and be tailored based on the immune profile of the patient. Future studies with larger arrays of cytokine profiles may provide more sensitive and specific modes of diagnostics in determining etiology of psychiatric conditions and provide guidance in individual therapies. A better understanding of the pharmacological mechanism of current anti-inflammatory agents will help discover new therapeutic targets and drugs. This multimodal approach will ultimately foster the understanding of the biological basis of mental disorders and their interaction with the immune system. Despite the drawbacks highlighted by some meta-analyses, the preliminary results are very promising.

## References

1. Müller N. A brief history of immunological research into psychosis and pathways for immune influence of the brain. Curr Top Behav Neurosci. 2020;44:1–8.
2. J S MD. Psychoimmunology: implications for future research. South Med J. 1992;85(4): 388–96, 402.
3. Miller AH, Maletic V, Raison CL. Inflammation and its discontents: the role of cytokines in the pathophysiology of major depression. Biol Psychiatry. 2009;65(9):732–41.
4. Careaga M, Van de Water J, Ashwood P. Immune dysfunction in autism: a pathway to treatment. Neurotherapeutics. 2010;7(3):283–92.
5. Teixeira AL, Rodrigues DH, Marques AH, Miguel EC, Fontenelle LF. Searching for the immune basis of obsessive-compulsive disorder. Neuroimmunomodulation. 2014;21(2–3): 152–8.
6. Wang Z, Caughron B, Young MRI. Posttraumatic stress disorder: an immunological disorder? Front Psych. 2017;8:222.

7. Caso JR, Graell M, Navalón A, MacDowell KS, Gutiérrez S, Soto M, Leza JC, Carrasco JL, Marsá MD. Dysfunction of inflammatory pathways in adolescent female patients with anorexia nervosa. Prog Neuropsychopharmacol Biol Psychiatry. 2020;96:109727.
8. Appleton J. The gut-brain axis: influence of microbiota on mood and mental health. Integr Med (Encinitas). 2018;17(4):28–32.
9. Vollhardt LT. Psychoneuroimmunology: a literature review. Am J Orthopsychiatry. 1991;61 (1):35–47.
10. Pape K, Tamouza R, Leboyer M, Zipp F. Immunoneuropsychiatry—novel perspectives on brain disorders. Nat Rev Neurol. 2019;15(6):317–28.
11. Colpo GD, Leboyer M, Dantzer R, Trivedi MH, Teixeira AL. Immune-based strategies for mood disorders: facts and challenges. Expert Rev Neurother. 2018;18(2):139–52.
12. Li M, Soczynska JK, Kennedy SH. Inflammatory biomarkers in depression: an opportunity for novel therapeutic interventions. Curr Psychiatry Rep. 2011;13(5):316–20.
13. Müller N, Myint AM, Schwarz MJ. Inflammatory biomarkers and depression. Neurotox rex. 2011;19:308–18.
14. Mellins CA, Malee KM. Understanding the mental health of youth living with perinatal HIV infection: lessons learned and current challenges. J Int AIDS Soc. 2013;16(1):18593.
15. Omdal R, Husby G, Mellgren SI. Mental health status in systemic lupus erythematosus. Scand J Rheumatol. 1995;24(3):142–5.
16. Roh JS, Sohn DH. Damage-associated molecular patterns in inflammatory diseases. Immune Netw. 2018;18(4):e27.
17. Kelley KW, Kent S. The legacy of sickness behaviors. Front Psych. 2020;11:607269.
18. Bennett JM, Reeves G, Billman GE, Sturmberg JP. Inflammation-nature's way to efficiently respond to all types of challenges: implications for understanding and managing "the epidemic" of chronic diseases. Front Med (Lausanne). 2018;5:316.
19. Lucas SM, Rothwell NJ, Gibson RM. The role of inflammation in CNS injury and disease. Br J Pharmacol. 2006;147 Suppl 1(Suppl 1):S232–40.
20. Antel JP, Becher B, Ludwin SK, Prat A, Quintana FJ. Glial cells as regulators of neuroimmune interactions in the central nervous system. J Immunol. 2020;204(2):251–5.
21. Qin Y, Wang N, Zhang X, Han X, Zhai X, Lu Y. IDO and TDO as a potential therapeutic target in different types of depression. Metab Brain Dis. 2018;33(6):1787–800.
22. Brás JP, Pinto S, Almeida MI, Prata J, von Doellinger O, Coelho R, Barbosa MA, Santos SG. Peripheral biomarkers of inflammation in depression: evidence from animal models and clinical studies. Methods Mol Biol. 2019;2011:467–92.
23. Toft H, Lien L, Neupane SP, Abebe DS, Tilden T, Wampold BE, Bramness JG. Cytokine concentrations are related to level of mental distress in inpatients not using anti-inflammatory drugs. Acta Neuropsychiatr. 2020;32(1):23–31.
24. Terrando N, Monaco C, Ma D, Foxwell BMJ, Feldman M, Maze M. Tumor necrosis factor-alpha triggers a cytokine cascade yielding postoperative cognitive decline. Proc Natl Acad Sci U S A. 2010;107:20518–22.
25. Meneses G, Bautista M, Florentino A, Díaz G, Acero C, Besedovsky H, Meneses D, Feury A, Del Rey A, Gevorkian G, Fragoso G, Sciutto E. Electric stimulation of the vagus nerve reduced mouse neuroinflammation induced by lipopolysaccharide. J Inflamm. 2016;13:33.
26. Silverman MN, Pearce BD, Biron CA, Miller AH. Immune modulation of the hypothalamic-pituitary-adrenal (HPA) axis during viral infection. Viral Immunol. 2005;18(1):41–78.
27. Hughes HK, Ashwood P. Overlapping evidence of innate immune dysfunction in psychotic and affective disorders. Brain Behav Immun. 2020;2:100038.
28. Rao J, Harry G, Rapoport SK, HW. Increased excitotoxicity and neuroinflammatory markers in postmortem frontal cortex from bipolar disorder patients. Mol Psychiatry. 2010;15:384–92.
29. Chrousos GP. Stress, chronic inflammation, and emotional and physical well-being: concurrent effects and chronic sequelae. J Allergy Clin Immunol. 2000;106(5 Suppl):S275–91.

30. Baumeister D, Akhtar R, Ciufolini S, Pariante CM, Mondelli V. Childhood trauma and adulthood inflammation: a meta-analysis of peripheral C-reactive protein, interleukin-6 and tumour necrosis factor-α. Mol Psychiatry. 2016;21:642–9.
31. Johnson JD, Barnard DF, Kulp AC, Mehta DM. Neuroendocrine regulation of brain cytokines after psychological stress. J Endocr Soc. 2019;3(7):1302–20.
32. Mithaiwala MN, Santana-Coelho D, Porter GA, O'Connor JC. Neuroinflammation and the kynurenine pathway in CNS disease: molecular mechanisms and therapeutic implications. Cell. 2021;10(6):1548.
33. Zhu CB, Blakely R, Hewlett W. The proinflammatory cytokines interleukin-1beta and tumor necrosis factor-alpha activate serotonin transporters. Neuropsychopharmacology. 2006;31: 2121–31.
34. Hovatta I, Juhila J, Donner J. Oxidative stress in anxiety and comorbid disorders. Neurosci Res. 2010;68(4):261–75.
35. Rooney S, Sah A, Unger MS, Kharitonova M, Sartori SB, Schwarcer C, Aigner L, Kettenmann H, Wolf SA, Singewald N. Neuroinflammatory alterations in trait anxiety: modulatory effects of minocycline. Transl Psychiatry. 2020;10:256.
36. Murphy TK, Petitto JM, Voeller KK, Goodman WK. Obsessive compulsive disorder: is there an association with childhood streptococcal infections and altered immune function? Semin Clin Neuropsychiatry. 2001;6(4):266–76.
37. Orefici G, Cardona F, Cox CJ, Cunningham MW, Ferreti JJ, Stevens DL, Fischetti VA. Pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections (PANDAS) 2016. In: Ferretti JJ, Stevens DL, Fischetti VA, editors. Streptococcus pyogenes: basic biology to clinical manifestations [internet]. Oklahoma City (OK): University of Oklahoma Health Sciences Center; 2016. Available from: https://www.ncbi.nlm.nih.gov/books/NBK333433/.
38. Murphy TK, Patel PD, McGuire JF, Kennel A, Mutch PJ, Parker-Athill EC, Hanks CE, Lewin AB, Storch EA, Toufexis MD, Dadlani GH, Rodriguez CA. Characterization of the pediatric acute-onset neuropsychiatric syndrome phenotype. J Child Adolesc Psychopharmacol. 2015;25(1):14–25.
39. Zibordi F, Zorzi G, Carecchio M, Nardocci N. CANS: childhood acute neuropsychiatric syndromes. Eur J Paediatr Neurol. 2018;22(2):316–20.
40. Orlovska S, Vestergaard CH, Bech BH, Nordentoft M, Vestergaard M, Benros ME. Association of streptococcal throat infection with mental disorders: testing key aspects of the PANDAS hypothesis in a nationwide study. JAMA Psychiat. 2017;74(7):740–6.
41. Gray SM, Bloch MH. Systematic review of proinflammatory cytokines in obsessive-compulsive disorder. Curr Psychiatry Rep. 2012;14(3):220–8.
42. Rodríguez N, Morer A, González-Navarro EA, Serra-Pages C, Boloc D, Torres T, García-Cerro S, Mas S, Gassó P, Lázaro L. Inflammatory dysregulation of monocytes in pediatric patients with obsessive-compulsive disorder. J Neuroinflammation. 2017;14(1):261.
43. Marazziti D, Mucci F, Fontenelle LF. Immune system and obsessive-compulsive disorder. Psychoneuroendocrinology. 2018;93:39–44.
44. Bookwalter DB, Roenfeldt KA, LeardMann CA, Kong SY, Riddle MS, Rull RP. Posttraumatic stress disorder and risk of selected autoimmune diseases among US military personnel. BMC Psychiatry. 2020;20(1):23.
45. Almli LM, Fani N, Smith AK, Ressler KJ. Genetic approaches to understanding post-traumatic stress disorder. Int J Neuropsychopharmacol. 2014;17(2):355–70.
46. Gola H, Engler H, Sommershof A, Adenauer H, Kolassa S, Schedlowski M, Goettrup M, elbert T, Kolassa I. Posttraumatic stress disorder is associated with an enhanced spontaneous production of pro-inflammatory cytokines by peripheral blood mononuclear cells. BMC Psychiatry. 2013;13:40.
47. Hori H, Kim Y. Inflammation and post-traumatic stress disorder. Psychiatry Clin Neurosci. 2019;73(4):143–53.

48. Raison CL, Miller AH. Is depression an inflammatory disorder? Curr Psychiatry Rep. 2011;13 (6):467–75.
49. Halaris A. Inflammation and depression but where does the inflammation come from? Curr Opin Psychiatry. 2019;32(5):422–8.
50. Nobis A, Zalewski D, Waszkiewicz N. Peripheral markers of depression. J Clin Med. 2020;9 (12):3793.
51. Benros ME, Waltoft BL, Nordentoft M, Ostergaard SD, Eaton WW, Krogh J, Mortensen PB. Autoimmune diseases and severe infections as risk factors for mood disorders: a nationwide study. JAMA Psychiat. 2013;70(8):812–20.
52. Capuron L, Hauser P, Hinze-Selch D, Miller AH, Neveu PJ. Treatment of cytokine-induced depression. Brain Behav Immun. 2002;16(5):575–80.
53. Kenis G, Maes M. Effects of antidepressants on the production of cytokines. Int J Neuropsychopharmacol. 2002;5(4):401–12.
54. Arteaga-Henríquez G, Simon MS, Burger B, Weidinger E, Wijkhuijs A, Arolt V, Birkenhager TK, Musil R, Müller N, Drexhage HA. Low-grade inflammation as a predictor of antidepressant and anti-inflammatory therapy response in MDD patients: a systematic review of the literature in combination with an analysis of experimental data collected in the EU-MOODINFLAME consortium. Front Psych. 2019;10:458.
55. Miller AH, Raison CL. The role of inflammation in depression: from evolutionary imperative to modern treatment target. Nat Rev Immunol. 2016;16(1):22–34.
56. Sakurai M, Yamamoto Y, Kanayama N, Hasegawa M, Mouri A, Takemura M, Matsunami H, Miyauchi T, Tokura T, Kimura H, Ito M, Umemura E, Boku AS, Nagashima W, Tonoike T, Kurita K, Ozaki N, Nabeshima T, Saito K. Serum metabolic profiles of the tryptophan-kynurenine pathway in the high risk subjects of major depressive disorder. Sci Rep. 2020;10 (1):1961.
57. Kaestner F, Hettich M, Peters M, Sibrowski W, Hetzel G, Ponath G, Arolt V, Cassens U, Rothermundt M. Different activation patterns of pro-inflammatory cytokines in melancholic and non-melancholic major depression are associated with HPA axis activity. J Affect Disord. 2005;87(2–3):305–11.
58. Vreeburg SA, Hoogendijk WJ, van Pelt J, Derijk RH, Verhagen JC, van Dyck R, Smit JH, Zitman FG, Penninx BW. Major depressive disorder and hypothalamic-pituitary-adrenal axis activity: results from a large cohort study. Arch Gen Psychiatry. 2009;66(6):617–26.
59. Cain D, Cidlowski J. Immune regulation by glucocorticoids. Nat Rev Immunol. 2017;17:233–47.
60. Rosenblat JD, McIntyre RS. Bipolar disorder and immune dysfunction: epidemiological findings, proposed pathophysiology and clinical implications. Brain Sci. 2017;7(11):144.
61. Wang AK, Miller BJ. Meta-analysis of cerebrospinal fluid cytokine and tryptophan catabolite alterations in psychiatric patients: comparisons between schizophrenia, bipolar disorder, and depression. Schizophr Bull. 2018;44(1):75–83.
62. Wieck A, Grassi-Oliveira R, do Prado CH, Rizzo LB, de Oliveira AS, Kommers-Molina J, Viola TW, Marciano Vieira EL, Teixeira AL, Bauer ME. Pro-inflammatory cytokines and soluble receptors in response to acute psychosocial stress: differential reactivity in bipolar disorder. Neurosci Lett. 2014;580:17–21.
63. Allende LM, Corell A, Manzanares J, Madruga D, Marcos A, Madroño A, López-Goyanes A, García-Pérez MA, Moreno JM, Rodrigo M, Sanz F, Arnaiz-Villena A. Immunodeficiency associated with anorexia nervosa is secondary and improves after refeeding. Immunology. 1998;94(4):543–51.
64. Słotwińska SM, Słotwiński R. Immune disorders in anorexia. Cent Eur J Immunol. 2017;42 (3):294–300.
65. Mustafa A, Ward A, Treasure J, Peakman M. T lymphocyte subpopulations in anorexia nervosa and refeeding. Clin Immunol Immunopathol. 1997;82(3):282–9.

66. Nilsson IAK, Millischer V, Göteson A, Hübel C, Thornton LM, Bulik CM, Schalling M, Landén M. Aberrant inflammatory profile in acute but not recovered anorexia nervosa. Brain Behav Immun. 2020;88:718–24.
67. Bou Khalil R, de Muylder O, Hebborn FL. Treatment of anorexia nervosa with TNF-α down-regulating agents. Eat Weight Disord. 2011;16(4):e300.
68. Comer AL, Carrier M, Tremblay MÈ, Cruz-Martín A. The inflamed brain in schizophrenia: the convergence of genetic and environmental risk factors that lead to uncontrolled neuroinflammation. Front Cell Neurosci. 2020;14:274.
69. García-Bueno B, Bioque M, MacDowell KS, Santabárbara J, Martínez-Cengotitabengoa M, Moreno C, Sáiz PA, Berrocoso E, Gassó P, Fe Barcones M, González-Pinto A, Parellada M, Bobes J, Micó JA, Bernardo M, Leza JC, FLAMM-PEPs study, Centro de Investigación Biomédica en red de Salud Mental (CIBERSAM), Spain. Pro−/anti-inflammatory dysregulation in early psychosis: results from a 1-year follow-up study. Int J Neuropsychopharmacol. 2014;18(2):pyu037.
70. Jenkins TA. Perinatal complications and schizophrenia: involvement of the immune system. Front Neurosci. 2013;7:110.
71. Smith SEP, Li J, Garbett K, Mirnics K, Patterson PH. Maternal immune activation alters fetal brain development through interleukin-6. J Neurosci. 2007;27(40):10695–702.
72. Schmitt A, Bertsch T, Tost H, Bergmann A, Henning U, Klimke A, Falkai P. Increased serum interleukin-1beta and interleukin-6 in elderly, chronic schizophrenic patients on stable anti-psychotic medication. Neuropsychiatr Dis Treat. 2005;1(2):171–7.
73. Momtazmanesh S, Zare-Shahabadi A, Rezaei N. Cytokine alterations in schizophrenia: an updated review. Front Psych. 2019;10:892.
74. Yang AC, Tsai SJ. New targets for schizophrenia treatment beyond the dopamine hypothesis. Int J Mol Sci. 2017;18(8):1689.
75. Müller N. Inflammation in schizophrenia: pathogenetic aspects and therapeutic considerations. Schizophr Bull. 2018;44(5):973–82.
76. Reale M, Costantini E, Greig NH. Cytokine imbalance in schizophrenia. From research to clinic: potential implications for treatment. Front Psych. 2021;12:536257.
77. Meltzer A, Van de Water J. The role of the immune system in autism spectrum disorder. Neuropsychopharmacology. 2017;42(1):284–98.
78. Chen SW, Zhong XS, Jiang LN, Zheng XY, Xiong YQ, Ma SJ, Qiu M, Huo ST, Ge J, Chen Q. Maternal autoimmune diseases and the risk of autism spectrum disorders in offspring: a systematic review and meta-analysis. Behav Brain Res. 2016;296:61–9.
79. Keil A, Daniels JL, Forssen U, Hultman C, Cnattingius S, Söderberg KC, Feychting M, Sparen P. Parental autoimmune diseases associated with autism spectrum disorders in offspring. Epidemiology. 2010;21(6):805–8.
80. Masi A, Glozier N, Dale R, Guastella AJ. The immune system, cytokines, and biomarkers in autism spectrum disorder neurosci. Dengue Bull. 2017;33:194–204.
81. Dantzer R. Neuroimmune interactions: from the brain to the immune system and vice versa. Physiol Rev. 2018;98(1):477–504.
82. Kopschina Feltes P, Doorduin J, Klein HC, Juárez-Orozco LE, Dierckx RA, Moriguchi-Jeckel CM, de Vries EF. Anti-inflammatory treatment for major depressive disorder: implications for patients with an elevated immune profile and non-responders to standard antidepressant therapy. J Psychopharmacol. 2017;31(9):1149–65.
83. Miller AH, Raison CL. Are anti-inflammatory therapies viable treatments for psychiatric disorders?: where the rubber meets the road. JAMA Psychiat. 2015;72(6):527–8.
84. Gałecki P, Mossakowska-Wójcik J, Talarowska M. The anti-inflammatory mechanism of antidepressants—SSRIs, SNRIs. Prog Neuropsychopharmacol Biol Psychiatry. 2018;80 (Pt C):291–4.
85. Berk M, Dean O, Drexhage H, McNeil JJ, Moylan S, O'Neil A, Davey CG, Sanna L, Maes M. Aspirin: a review of its neurobiological properties and therapeutic potential for mental illness. BMC Med. 2013;11:74.

86. Janelidze S, Brundin L. Inflammation in suicidality: implications for novel treatment options. Mod Trends Pharmacopsychiatry. 2013;28:188–202.
87. Sommer IE, van Westrhenen R, Begemann MJ, de Witte LD, Leucht S, Kahn RS. Efficacy of anti-inflammatory agents to improve symptoms in patients with schizophrenia: an update. Schizophr Bull. 2014;40:181–91.
88. Köhler-Forsberg O, Lydholm CN, Hjorthøj C, Nordentoft M, Mors O, Benros ME. Efficacy of anti-inflammatory treatment on major depressive disorder or depressive symptoms: meta-analysis of clinical trials. Acta Psychiatr Scand. 2019;139(5):404–19.
89. Hong J, Bang M. Anti-inflammatory strategies for schizophrenia: a review of evidence for therapeutic applications and drug repurposing. Clin Psychopharmacol Neurosci. 2020;18(1): 10–24.
90. Jeppesen R, Christensen RHB, Pedersen EMJ, Nordentoft M, Hjorthøj C, Köhler-Forsberg O, Benros ME. Efficacy and safety of anti-inflammatory agents in treatment of psychotic disorders—a comprehensive systematic review and meta-analysis. Brain Behav Immun. 2020;90:364–80.
91. Pereira AC, Oliveira J, Silva S, Madeira N, Pereira CMF, Cruz MT. Inflammation in bipolar disorder (BD): identification of new therapeutic targets. Pharmacol Res. 2021;163:105325.
92. Miller BJ, Buckley PF. The case for adjunctive monoclonal antibody immunotherapy in schizophrenia. Psychiatr Clin North Am. 2016;39(2):187–98.
93. Uzzan S, Azab AN. Anti-TNF-α compounds as a treatment for depression. Molecules. 2021;26(8):2368.
94. Essali N, Goldsmith DR, Carbone L, Miller BJ. Psychosis as an adverse effect of monoclonal antibody immunotherapy. Brain Behav Immun. 2019;81:646–9.
95. Grüber L, Bunse T, Weidinger E, Reichard H, Müller N. Adjunctive recombinant human interferon gamma-1b for treatment-resistant schizophrenia in 2 patients. J Clin Psychiatry. 2014;75(11):1266–7.
96. Prather AA, Rabinovitz M, Pollock BG, Lotrich FE. Cytokine-induced depression during IFN-alpha treatment: the role of IL-6 and sleep quality. Brain Behav Immun. 2009;23(8): 1109–16.
97. Rhen T, Cidlowski JA. Antiinflammatory action of glucocorticoids—new mechanisms for old drugs. N Engl J Med. 2005;353(16):1711–23.
98. Chakraborty S, Pramanik J, Mahata B. Revisiting steroidogenesis and its role in immune regulation with the advanced tools and technologies. Genes Immun. 2021;22:125–40.
99. Pivonello R, Simeoli C, De Martino MC, Cozzolino A, De Leo M, Iacuaniello D, Pivonello C, Negri M, Pellecchia MT, Iasevoli F, Colao A. Neuropsychiatric disorders in Cushing's syndrome. Front Neurosci. 2015;9:129.
100. Mrakotsky C, Forbes PW, Bernstein JH, Grand RJ, Bousvaros A, Szigethy E, Waber DP. Acute cognitive and behavioral effects of systemic corticosteroids in children treated for inflammatory bowel disease. J Int Neuropsychol Soc. 2013;19(1):96–109.
101. Hill AR, Spencer-Segal JL. Glucocorticoids and the brain after critical illness. Endocrinology. 2021;162(3):bqaa242.
102. Qin D, Li Z, Li Z, Wang L, Hu Z, Lü L, Wang Z, Liu Y, Yin Y, Li Z, Hu X. Chronic glucocorticoid exposure induces depression-like phenotype in rhesus macaque (Macaca Mulatta). Front Neurosci. 2019;13:188.
103. Frank MG, Watkins LR, Maier SF. Stress- and glucocorticoid-induced priming of neuroinflammatory responses: potential mechanisms of stress-induced vulnerability to drugs of abuse. Brain Behav Immun. 2011;25 Suppl 1(Suppl 1):S21–8.
104. Holsboer F. The corticosteroid receptor hypothesis of depression. Neuropsychopharmacology. 2000;23:477–501.
105. Szeszko PR, Lehrner A, Yehuda R. Glucocorticoids and hippocampal structure and function in PTSD. Harv Rev Psychiatry. 2018;26(3):142–57.
106. de Quervain D, Wolf OT, Roozendaal B. Glucocorticoid-induced enhancement of extinction-from animal models to clinical trials. Psychopharmacology (Berl). 2019;236(1):183–99.

107. de Quervain DJ, Margraf J. Glucocorticoids for the treatment of post-traumatic stress disorder and phobias: a novel therapeutic approach. Eur J Pharmacol. 2008;583(2–3):365–71.
108. Bavarsad Shahripour R, Harrigan MR, Alexandrov AV. N-acetylcysteine (NAC) in neurological disorders: mechanisms of action and therapeutic opportunities. Brain Behav. 2014;4(2): 108–22.
109. Deepmala SJ, Kumar N, Delhey L, Berk M, Dean O, Spielholz C, Frye R. Clinical trials of N-acetylcysteine in psychiatry and neurology: a systematic review. Neurosci Biobehav Rev. 2015;55:294–321.
110. Willborn RJ, Hall CP, Fuller MA. Recycling N-acetylcysteine: a review of evidence for adjunctive therapy in schizophrenia. Ment Health Clin. 2019;9(3):116–23.
111. Ooi SL, Green R, Pak SC. N-Acetylcysteine for the treatment of psychiatric disorders: a review of current evidence. Biomed Res Int. 2018;2018:2469486.
112. Pae CU, Marks DM, Han C, Patkar AA. Does minocycline have antidepressant effect? Biomed Pharmacother. 2008;62(5):308–11.
113. Zhang L, Zhao J. Profile of minocycline and its potential in the treatment of schizophrenia. Neuropsychiatr Dis Treat. 2014;10:1103–11.
114. Romero-Miguel D, Lamanna-Rama N, Casquero-Veiga M, Gómez-Rangel V, Desco M, Soto-Montenegro ML. Minocycline in neurodegenerative and psychiatric diseases: an update. Eur J Neurol. 2021;28(3):1056–81.
115. Nettis MA, Lombardo G, Hastings C, Zajkowska Z, Mariani N, Nikkheslat N, Worrell C, Enache D, McLaughlin A, Kose M, Sforzini L, Bogdanova A, Cleare A, Young AH, Pariante CM, Mondelli V. Augmentation therapy with minocycline in treatment-resistant depression patients with low-grade peripheral inflammation: results from a double-blind randomised clinical trial. Neuropsychopharmacology. 2021;46(5):939–48.
116. Husain MI, Chaudhry IB, Hamirani MM, Minhas FA, Kazmi A, Hodsoll J, Haddad PM, Deakin JF, Husain N, Young AH. Minocycline and celecoxib as adjunctive treatments for bipolar depression: a study protocol for a multicenter factorial design randomized controlled trial. Neuropsychiatr Dis Treat. 2016;13:1–8.
117. Chan SY, Capitão L, Probert F, Klinge C, Hoeckner S, Harmer CJ, Cowen PJ, Anthony DC, Burnet PWJ. A single administration of the antibiotic, minocycline, reduces fear processing and improves implicit learning in healthy volunteers: analysis of the serum metabolome. Transl Psychiatry. 2020;10(1):148.
118. Jain M, Ridke P. Anti-inflammatory effects of statins: clinical evidence and basic mechanisms. Nat Rev Drug Discov. 2005;4:977–87.
119. Kim SW, Kang HJ, Jhon M, Kim JW, Lee JY, Walker AJ, Agustini B, Kim JM, Berk M. Statins and inflammation: new therapeutic opportunities in psychiatry. Front Psych. 2019;10:103.
120. Pasco JA, Jacka FN, Williams LJ, Henry MJ, Nicholson GC, Kotowicz MA, Berk M. Clinical implications of the cytokine hypothesis of depression: the association between use of statins and aspirin and the risk of major depression. Psychother Psychosom. 2010;9:323–5.
121. Cham S, Koslik HJ, Golomb BA. Mood, personality, and behavior changes during treatment with statins: a case series. Drug Saf Case Rep. 2016;3(1):1.
122. Shen H, Li R, Yan R, Zhou X, Feng X, Zhao M, Xiao H. Adjunctive therapy with statins in schizophrenia patients: a meta-analysis and implications. Psychiatry Res. 2018;262:84–93.
123. Lang KW. Omega-3 fatty acids and mental health. Global Health J. 2020;4:18–30.
124. Robinson DG, Gallego JA, John M, Hanna LA, Zhang JP, Birnbaum ML, Greenberg J, Naraine M, Peters BD, McNamara RK, Malhotra AK, Szeszko PR. A potential role for adjunctive omega-3 polyunsaturated fatty acids for depression and anxiety symptoms in recent onset psychosis: results from a 16 week randomized placebo-controlled trial for participants concurrently treated with risperidone. Schizophr Res. 2019;204:295–303.
125. Ross BM, Seguin J, Sieswerda LE. Omega-3 fatty acids as treatments for mental illness: which disorder and which fatty acid? Lipids Health Dis. 2007;6:21.

126. Königs A, Kiliaan AJ. Critical appraisal of omega-3 fatty acids in attention-deficit/hyperactivity disorder treatment. Neuropsychiatr Dis Treat. 2016;12:1869–82.
127. Hsiao EY, McBride SW, Hsien S, Sharon G, Hyde ER, McCue T, Codelli JA, Chow J, Reisman SE, Petrosino JF, Patterson PH, Mazmanian SK. Microbiota modulate behavioral and physiological abnormalities associated with neurodevelopmental disorders. Cell. 2013;155(7):1451–63.
128. Huang R, Wang K, Hu J. Effect of probiotics on depression: a systematic review and meta-analysis of randomized controlled trials. Nutrients. 2016;8(8):483.
129. Rao P, Knaus EE. Evolution of nonsteroidal anti-inflammatory drugs (NSAIDs): cyclooxygenase (COX) inhibition and beyond. J Pharm Pharm Sci. 2008;11(2):81s–110s.
130. Smith WL, Urade Y, Jakobsson PJ. Enzymes of the cyclooxygenase pathways of prostanoid biosynthesis. Chem Rev. 2011;111(10):5821–65.
131. Kaufmann WE, Andreasson KI, Isakson PC, Worley PF. Cyclooxygenases and the central nervous system. Prostaglandins. 1997;54(3):601–24.
132. López DE, Ballaz SJ. The role of brain cyclooxygenase-2 (Cox-2) beyond neuroinflammation: neuronal homeostasis in memory and anxiety. Mol Neurobiol. 2020;57(12):5167–76.
133. Mirjany M, Ho L, Pasinetti GM. Role of cyclooxygenase-2 in neuronal cell cycle activity and glutamate-mediated excitotoxicity. J Pharmacol Exp Ther. 2002;301(2):494–500.
134. Minghetti L. Cyclooxygenase-2 (COX-2) in inflammatory and degenerative brain diseases. J Neuropathol Exp Neurol. 2004;63:901–10.
135. Yokota O, Terada S, Ishihara T, Nakashima H, Kugo A, Ujike H, Tsuchiya K, Ikeda K, Saito Y, Murayama S, Ishizu H, Kuroda S. Neuronal expression of cyclooxygenase-2, a pro-inflammatory protein, in the hippocampus of patients with schizophrenia. Prog Neuropsychopharmacol Biol Psychiatry. 2004;28(4):715–21.
136. Bavaresco DV, da Rosa MI, Uggioni MLR, Ferraz SD, Pacheco TR, Toé HCZD, da Silveira AP, Quadros LFA, de Souza TD, Varela RB, Vieira AAS, Pizzol FD, Valvassori SS, Quevedo J. Increased inflammatory biomarkers and changes in biological rhythms in bipolar disorder: a case-control study. J Affect Disord. 2020;271:115–22.
137. Parepally JM, Mandula H, Smith QR. Brain uptake of nonsteroidal anti-inflammatory drugs: ibuprofen, flurbiprofen, and indomethacin. Pharm Res. 2006;23(5):873–81.
138. Marini S, De Berardis D, Vellante F, Santacroce R, Orsolini L, Valchera A, Girinelli G, Carano A, Fornaro M, Gambi F, Martinotti G, Di Giannantonio M. Celecoxib adjunctive treatment to antipsychotics in schizophrenia: a review of randomized clinical add-on trials. Mediators Inflamm. 2016;2016:3476240.
139. Müller N. COX-2 inhibitors, aspirin, and other potential anti-inflammatory treatments for psychiatric disorders. Front Psych. 2019;10:375.
140. Sethi R, Gómez-Coronado N, Walker AJ, Robertson OD, Agustini B, Berk M, Dodd S. Neurobiology and therapeutic potential of cyclooxygenase-2 (COX-2) inhibitors for inflammation in neuropsychiatric disorders. Front Psych. 2019;10:605.
141. Weiser M, Zamora D, Levi L, Nastas I, Gonen I, Radu P, Matei V, Nacu A, Boronin L, Davidson M, Davis JM. Adjunctive aspirin vs placebo in patients with schizophrenia: results of two randomized controlled trials. Schizophr Bull. 2021;47(4):1007–87.
142. Müller N, Schwarz MJ. COX-2 inhibition in schizophrenia and major depression. Curr Pharm Des. 2008;14(14):1452–65.
143. Zheng W, Cai DB, Yang XH, Ungvari GS, Ng CH, Müller N, Ning YP, Xiang YT. Adjunctive celecoxib for schizophrenia: a meta-analysis of randomized, double-blind, placebo-controlled trials. J Psychiatr Res. 2017;92:139–46.
144. Iyengar RL, Gandhi S, Aneja A, Thorpe K, Razzouk L, Greenberg J, Mosovich S, Farkouh ME. NSAIDs are associated with lower depression scores in patients with osteoarthritis. Am J Med. 2013;126(11):1017.e11–8.
145. Lehrer S, Rheinstein PH. Nonsteroidal anti-inflammatory drugs (NSAIDs) reduce suicidal ideation and depression. Discov Med. 2019;28(154):205–12.

146. Perrone MG, Centonze A, Miciaccia M, Ferorelli S, Scilimati A. Cyclooxygenase inhibition safety and efficacy in inflammation-based psychiatric disorders. Molecules. 2020;25(22): 5388.
147. Gallagher PJ, Castro V, Fava M, Weilburg JB, Murphy SN, Gainer VS, Churchill SE, Kohane IS, Iosifescu DV, Smoller JW, Perlis RH. Antidepressant response in patients with major depression exposed to NSAIDs: a pharmacovigilance study. Am J Psychiatry. 2012;169(10): 1065–72.
148. Eyre HA, Air T, Proctor S, Rositano S, Baune BT. A critical review of the efficacy of non-steroidal anti-inflammatory drugs in depression. Prog Neuropsychopharmacol Biol Psychiatry. 2015;57:11–6.
149. Dembo G, Park SB, Kharasch ED. Central nervous system concentrations of cyclooxygenase-2 inhibitors in humans. Anesthesiology. 2005;102(2):409–15.
150. Müller N, Schwarz MJ, Dehning S, Douhe A, Cerovecki A, Goldstein-Müller B, Spellmann I, Hetzel G, Maino K, Kleindienst N, Möller HJ, Arolt V, Riedel M. The cyclooxygenase-2 inhibitor celecoxib has therapeutic effects in major depression: results of a double-blind, randomized, placebo controlled, add-on pilot study to reboxetine. Mol Psychiatry. 2006;11(7): 680–4.
151. Akhondzadeh S, Tabatabaee M, Amini H, Ahmadi Abhari SA, Abbasi SH, Behnam B. Celecoxib as adjunctive therapy in schizophrenia: a double-blind, randomized and placebo-controlled trial. Schizophr Res. 2007;90(1–3):179–85.
152. Faridhosseini F, Sadeghi R, Farid L, Pourgholami M. Celecoxib: a new augmentation strategy for depressive mood episodes. A systematic review and meta-analysis of randomized placebo-controlled trials. Hum Psychopharmacol. 2014;29(3):216–23.
153. Na KS, Lee KJ, Lee JS, Cho YS, Jung HY. Efficacy of adjunctive celecoxib treatment for patients with major depressive disorder: a meta-analysis. Prog Neuropsychopharmacol Biol Psychiatry. 2014;48:79–85. Erratum in: Prog Neuropsychopharmacol Biol Psychiatry. 2016;66:136.
154. Shalbafan M, Mohammadinejad P, Shariat SV, Alavi K, Zeinoddini A, Salehi M, Askari N, Akhondzadeh S. Celecoxib as an adjuvant to fluvoxamine in moderate to severe obsessive-compulsive disorder: a double-blind, placebo-controlled, randomized trial. Pharmacopsychiatry. 2015;48(4–5):136–40.
155. Halaris A, Cantos A, Johnson K, Hakimi M, Sinacore J. Modulation of the inflammatory response benefits treatment-resistant bipolar depression: a randomized clinical trial. J Affect Disord. 2020;261:145–52.
156. Fourrier C, Sampson E, Mills NT, Baune BT. Anti-inflammatory treatment of depression: study protocol for a randomised controlled trial of vortioxetine augmented with celecoxib or placebo. Trials. 2018;19(1):447.
157. Asadabadi M, Mohammadi MR, Ghanizadeh A, Modabbernia A, Ashrafi M, Hassanzadeh E, Forghani S, Akhondzadeh S. Celecoxib as adjunctive treatment to risperidone in children with autistic disorder: a randomized, double-blind, placebo-controlled trial. Psychopharmacology (Berl). 2013;225(1):51–9.
158. Çakici N, van Beveren NJM, Judge-Hundal G, Koola MM, Sommer IEC. An update on the efficacy of anti-inflammatory agents for patients with schizophrenia: a meta-analysis. Psychol Med. 2019;49(14):2307–19.
159. Müller N, Krause D, Dehning S, Musil R, Schennach-Wolff R, Obermeier M, Möller HJ, Klauss V, Schwarz MJ, Riedel M. Celecoxib treatment in an early stage of schizophrenia: results of a randomized, double-blind, placebo-controlled trial of celecoxib augmentation of amisulpride treatment. Schizophr Res. 2010;121(1–3):118–24.
160. Laan W, Grobbee DE, Selten JP, Heijnen CJ, Kahn RS, Burger H. Adjuvant aspirin therapy reduces symptoms of schizophrenia spectrum disorders: results from a randomized, double-blind, placebo-controlled trial. J Clin Psychiatry. 2010;71(5):520–7.
161. Hu K, Sjölander A, Lu D, Walker AK, Sloan EK, Fall K, Valdimarsdóttir U, Hall P, Smedby KE, Fang F. Aspirin and other non-steroidal anti-inflammatory drugs and depression, anxiety,

and stress-related disorders following a cancer diagnosis: a nationwide register-based cohort study. BMC Med. 2020;18(1):238.
162. Makunts T, Cohen IV, Lee KC, Abagyan R. Population scale retrospective analysis reveals distinctive antidepressant and anxiolytic effects of diclofenac, ketoprofen and naproxen in patients with pain. PLoS One. 2018;13(4):e0195521.
163. Leonard BE. Inflammation and depression: a causal or coincidental link to the pathophysiology? Acta Neuropsychiatr. 2018;30(1):1–16.
164. Fields C, Drye L, Vaidya V, Lyketsos C, ADAPT Research Group. Celecoxib or naproxen treatment does not benefit depressive symptoms in persons age 70 and older: findings from a randomized controlled trial. Am J Geriatr Psychiatry. 2012;20(6):505–13.
165. Baune BT. Are non-steroidal anti-inflammatory drugs clinically suitable for the treatment of symptoms in depression-associated inflammation? Curr Top Behav Neurosci. 2017;31:303–19.
166. Golia MT, Poggini S, Alboni S, Garofalo S, Ciano Albanese N, Viglione A, Ajmone-Cat MA, St-Pierre A, Brunello N, Limatola C, Branchi I, Maggi L. Interplay between inflammation and neural plasticity: both immune activation and suppression impair LTP and BDNF expression. Brain Behav Immun. 2019;81:484–94.
167. Aïd S, Bosetti F. Targeting cyclooxygenases-1 and -2 in neuroinflammation: therapeutic implications. Biochimie. 2011;93(1):46–51.
168. Schmid T, Brüne B. Prostanoids and resolution of inflammation—beyond the lipid-mediator class switch. Front Immunol. 2021;12:714042.
169. Hermanson DJ, Gamble-George JC, Marnett LJ, Patel S. Substrate-selective COX-2 inhibition as a novel strategy for therapeutic endocannabinoid augmentation. Trends Pharmacol Sci. 2014;35(7):358–67.
170. Tang B, Capitao C, Dean B, Thomas EA. Differential age- and disease-related effects on the expression of genes related to the arachidonic acid signaling pathway in schizophrenia. Psychiatry Res. 2012;196(2–3):201–6.
171. Kim HW, Rapoport SI, Rao JS. Altered arachidonic acid cascade enzymes in postmortem brain from bipolar disorder patients. Mol Psychiatry. 2011;16(4):419–28.
172. Bavaresco DV, Uggioni MLR, Simon CS, Colonetti T, Ferraz SD, Cruz MVB, Valvassori SS, Quevedo J, da Rosa MI. Evaluation of the arachidonic acid pathway in bipolar disorder: a systematic review. Mol Biol Rep. 2020;47(10):8209–17.
173. Su KP, Huang SY, Peng CY, Lai HC, Huang CL, Chen YC, Aitchison KJ, Pariante CM. Phospholipase A2 and cyclooxygenase 2 genes influence the risk of interferon-alpha-induced depression by regulating polyunsaturated fatty acids levels. Biol Psychiatry. 2010;67(6):550–7.
174. Rao JS, Rapoport SI. Mood-stabilizers target the brain arachidonic acid cascade. Curr Mol Pharmacol. 2009;2(2):207–14.
175. D'Acquisto F. Affective immunology: where emotions and the immune response converge. Dialogues Clin Neurosci. 2017;19(1):9–19.
176. Subramaniapillai M, Carmona NE, Rong C, McIntyre RS. Inflammation: opportunities for treatment stratification among individuals diagnosed with mood disorders. Dialogues Clin Neurosci. 2017;19(1):27–36.
177. Kroken RA, Sommer IE, Steen VM, Dieset I, Johnsen E. Constructing the immune signature of schizophrenia for clinical use and research; an integrative review translating descriptives into diagnostics. Front Psych. 2019;9:753.

178. Orlovska-Waast S, Köhler-Forsberg O, Brix SW, Nordentoft M, Kondziella D, Krogh J, Benros ME. Cerebrospinal fluid markers of inflammation and infections in schizophrenia and affective disorders: a systematic review and meta-analysis. Mol Psychiatry. 2019;24: 869–87.
179. Hidese S, Hattori K, Sasayama D, Tsumagari T, Miyakawa T, Matsumura R, Yokota Y, Ishida I, Matsuo J, Yoshida S, Ota M, Kunugi H. Cerebrospinal fluid inflammatory cytokine levels in patients with major psychiatric disorders: a multiplex immunoassay study. Front Pharmacol. 2021;11:594394.
180. Mondelli V, Vernon AC, Turkheimer F, Dazzan P, Pariante CM. Brain microglia in psychiatric disorders. Lancet Psychiatry. 2017;4(7):563–72.
181. McCoy MK, Tansey MG. TNF signaling inhibition in the CNS: implications for normal brain function and neurodegenerative disease. J Neuroinflammation. 2008;5:45.

# 21 The Glutamatergic System in Treatment-Resistant Depression and Comparative Effectiveness of Ketamine and Esketamine: Role of Inflammation?

Angelos Halaris and John Cook

**Abstract**

The glutamatergic system is the primary excitatory pathway within the CNS and is responsible for cognition, memory, learning, emotion, and mood. Because of its significant importance in widespread nervous system function, it is tightly regulated through multiple mechanisms, such as glutamate recycling, microglial interactions, and inflammatory pathways. Imbalance within the glutamatergic system has been implicated in a wide range of pathological conditions including neurodegenerative conditions, neuromuscular conditions, and mood disorders including depression. Major depressive disorder (MDD) is the most common mood disorder worldwide, has a high prevalence rate, and afflicts approximately 280 million people. While there are numerous treatments for the disease, 30–40% of patients are unresponsive to treatment and deemed treatment resistant; approximately another third experience only partial improvement (World Health Organization, Depression fact sheet [Internet], 2020). Esketamine, the S-enantiomer of ketamine, was approved by the Food and Drug Administration for treatment-resistant depression (TRD) in 2019 and has offered new hope to patients. It is the first treatment targeting the glutamatergic system through a complex mechanism. Numerous studies have implicated imbalance in the glutamatergic system in depression and treatment resistance. Esketamine and ketamine principally work through inhibition of the NMDA receptor, though more recent studies have implicated numerous other mechanisms mediating the antidepressant efficacy of these agents. These mechanisms include increase in brain-derived neurotrophic factor (BDNF), activation of mammalian target of the rapamycin complex

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(mTORC), and reduction in inflammation. Esketamine and ketamine have been shown to decrease inflammation in numerous ways principally through reducing pro-inflammatory cytokines (e.g., TNF-α, IL-6) (Loix et al., Acta Anaesthesiol Belg 62(1):47–58, 2011; Chen et al., Psychiatry Res 269:207–11, 2018; Kopra et al., J Psychopharmacol 35(8):934–45, 2021). This anti-inflammatory effect has also been shown to be involved in the antidepressive properties of both ketamine and esketamine (Chen et al., Psychiatry Res 269:207–11, 2018; Kopra et al., J Psychopharmacol 35(8):934–45, 2021).

**Keywords**

Major depressive disorder · Glutamatergic system · Inflammation · Cytokines · Ketamine · Esketamine · Treatment resistance

## 21.1 Introduction

This chapter aims to introduce the role of the glutamatergic system in treatment-resistant depression (TRD) including agents that target this system, such as ketamine and its enantiomers. Since its initial use as an antidepressant in clinical trials in 2000, ketamine has been a unique therapy due to its rapid onset of action and first-in-class mechanism targeting the glutamatergic system primarily through NMDA receptor inhibition [1]. Since the introduction of the monoamine hypothesis of depression in the 1960s, psychiatric research has established that a host of additional factors contribute to the pathophysiology of major depressive disorder (MDD) and treatment resistance [2, 3]. These include, but are not limited to, an upregulated HPA axis, metabolic dysfunction, microbiome composition, and inflammation. This chapter will (1) provide background into MDD and treatment-resistant depression (TRD) including its definition, prevalence, and treatment approaches; (2) highlight the role the glutamatergic system plays in depression; (3) highlight the use of ketamine and esketamine for TRD including benefits and drawbacks to therapy; and (4) describe the interactions between glutamatergic dysfunction, inflammation, and depression. In spite of the complexity and intricacy of these concepts, they represent an exciting new frontier in psychiatric diagnosis and treatment and overall, and ultimately, in achieving a higher rate of response and remission.

## 21.2 Background

Major depressive disorder (MDD) afflicts nearly 280 million people worldwide or approximately 3.8% of the world's population [4]. It is the most common psychiatric condition with severe morbidity including, but not limited to, overwhelming sadness and guilt, anhedonia, low energy, poor sleep and appetite, difficulty with concentration, loss of usual interests, weight gain or loss, and, most severely, suicide. Suicide

is currently the 17th leading cause of death worldwide and represents ~1.3% of all deaths globally [5, 6].

Treatments for MDD vary greatly with over 30 drugs across 5 different classes targeting serotonergic, noradrenergic, dopaminergic, and most recently glutamatergic systems. The development of most of these drugs was stimulated by the formulation of the monoamine hypothesis during the 1950s and 1960s. During that time, researchers were studying the role of serotonin and the hallucinogen lysergic acid diethylamide (LSD) [7–9]. Through this research, psychiatrists proposed a link between decreases in monoamines, particularly serotonin, norepinephrine, dopamine, and depression. The first treatment for depression was discovered accidentally while developing drugs for tuberculosis in the late 1950s. The drug, iproniazid, was originally marketed as an anti-TB drug but was commonly used off-label for MDD due to its ability to induce a state of euphoria [10]. Later it was discovered that iproniazid was a monoamine oxidase (MAO) inhibitor (MAOI) and thus became the first successful pharmacologic treatment for MDD [11]. MAO is a key enzyme in the breakdown of serotonin, norepinephrine, epinephrine, and dopamine [11]. With two isoforms, MAO-A and MAO-B, drugs can be specific for one of the enzymes or nonspecific. Inhibition of this enzyme results in an increase of these neurotransmitters in the CNS.

Today, additional mechanisms of commonly prescribed drugs include selective serotonin reuptake inhibitors (SSRIs), such as fluoxetine, sertraline, paroxetine, citalopram, escitalopram, and vilazodone; serotonin/norepinephrine reuptake inhibitors (SNRIs), such as duloxetine, desvenlafaxine, and levomilnacipran; tricyclic antidepressants (TCAs); or atypical antidepressants, such as bupropion.

Each of these drugs targets depression by increasing monoaminergic neurotransmission, particularly of serotonin, norepinephrine, and dopamine. More recently, however, the effect of these drugs has been suggested to be due, at least in part, to downstream effects on synaptic plasticity by increasing levels of numerous neurotrophins [12–15]. This may, at least partially, account for the delay of these drugs in achieving efficacy, even though a rise in monoamine levels may occur instantly.

The approval of esketamine for introduction into the market has introduced a novel mechanism of action for the treatment of depression. Esketamine, the S-enantiomer of ketamine, increases glutamate, the primary excitatory neurotransmitter in the CNS, through inhibition of the NMDA receptor on GABAergic neurons. This novel treatment approach offers new hope for the treatment-resistant patient population.

While there are numerous drugs to treat depression, a large percentage of patients are not adequately treated and experience persistent symptoms. It is estimated that approximately 50–60% of patients are unresponsive to one or more trials of antidepressants and 30–40% of all patients are nonresponders to at least two adequate treatments [16–18]. There is a significant debate as to what is considered treatment resistance; however, it is generally accepted to be failure of at least two adequate treatments, adequacy being defined as adequate dosing and length of treatment [16–18]. Once a patient is considered to have TRD, there are significantly

fewer treatment options. Today, there are only two pharmacotherapies approved for TRD including esketamine (Spravato) and olanzapine-fluoxetine combination (Symbyax). Outside of pharmacotherapies, other treatment modalities such as electroconvulsive therapy (ECT) and repetitive transcranial magnetic stimulation (rTMS) are also used.

The approval of esketamine in March of 2019 was particularly exciting for patients with TRD as it offers a fast-acting, novel mechanism of action through upregulation of the glutamatergic system. There is a significant ongoing investigation into the role of glutamate in depression, and the next section will summarize the current understanding.

## 21.3 Glutamate's Role in CNS Function

Although serotonin, norepinephrine, and dopamine were linked to depression as early as the 1960s, glutamate, the primary excitatory neurotransmitter of the CNS, was not implicated until the 1990s [19, 20]. In fact, glutamate was not even thought to be a neurotransmitter until the 1980s [19]. Up until that point, glutamate was viewed to be essential for brain metabolism as well as having general excitatory properties [21]. Since then, however, research into the role of glutamate has accelerated. It is now understood to be the most widespread neurotransmitter in the brain. Further, glutamate is now implicated in general cognition, learning, memory, and more recently emotion and mood regulation [22–25]. Glutamate is synthesized from glucose via the citric acid cycle or converted from glutamine by glutaminase [26]. Because of its widespread, critical function, it is one of the more tightly regulated systems within the brain. It is tightly regulated with numerous positive and negative feedback mechanisms. Additionally, after glutamate is released from the presynaptic terminal and binds to its receptors, it is quickly taken up by glutamate transporters on astrocytes. In astrocytes, it can be converted to glutamine via glutamine synthetase and recycled to the neuron. This tight regulation occurs in part due to significant tissue damage when glutamate is unbalanced (e.g., excitotoxicity). Imbalance within the glutamatergic system has been implicated in numerous pathologic states over the last two or three decades including neurodegenerative conditions, mood disorders, complications of stroke, and autism spectrum disorder among others [24, 27–30]. Mood disorders, such as depression, are now understood to entail glutamatergic imbalance, and this is presently an exciting new area of research.

## 21.4 Glutamate's Receptors

To better understand the role that glutamate plays in depression, it is important to discuss the diversity of glutamate receptors. Generally, there are two distinct types of glutamate receptors in the brain: ionotropic and metabotropic. Ionotropic receptors (iGluR) are ligand-gated ion channels that are nonselective cation channels allowing

the passage of $Na^+$, $K^+$, and $Ca^{2+}$ into the neuron [23, 31, 32]. These receptors lead to cellular excitation and include N-methyl-D-aspartate (NMDA) receptors, α-amino-3-hydroxyl-5-methyl-4-isoxazole-propionate (AMPA) receptors, and kainic acid receptors [23]. Metabotropic receptors (mGluR) are a family of receptors which eventually lead to ion channel opening through a signal cascade [32]. Many of these receptors are G-protein-coupled receptors which results in multiple steps to signaling. Activation of metabotropic receptors through glutamate binding results in a slower response and is linked to both increasing and decreasing excitability of the neuron. There are three distinct groups of metabotropic glutamate receptors in humans (i.e., group 1, group 2, group 3) with many receptors within each group (e.g., mGluR1, mGluR2) [32]. The colocalization of these receptors on synapses throughout the brain leads to a tremendous amount of diversity and complexity within the glutamatergic system. With this background, we can now move to discuss the current understanding of the role of the glutamatergic system in depression.

## 21.5 Glutamatergic System in Depression

Like many pathological mechanisms within the brain, the discovery of glutamate's role in depression was uncovered after the use of a therapeutic agent. In early rodent studies in the 1990s, animal models mimicked depression via stress-induced decrease in long-term potentiation. This means that by exposing the rodents to inescapable stress (e.g., forced swim test, tail suspension test), there is a decrease of neuronal efficiency in the hippocampus leading to a depressive-like state [20]. In essence, stress led to depression within the rodents by weakening connections throughout the brain. In these studies, administration of ketamine relieved this depressive-like episode by increasing rodents' urge to swim [20]. This led to the belief that glutamate played a role in depression. Unknown to the investigators at the time, it was later revealed that glutamate had been targeted as early as 1959 when cycloserine, a tuberculosis drug and partial NMDA antagonist, was shown to exert antidepressant effects [33].

The implication of glutamate playing a role in human depression has been largely attributed to experiments performed in 2000 [1]. At that point, Berman et al. used a sub-anesthetic dose of IV ketamine to improve depression in seven patients [1]. The study was groundbreaking because it showed that modulation of glutamate in the CNS leads to improvement in depression. Over the past two decades, research into the "glutamate hypothesis" has taken off. Studies in humans have been complicated and have shown a muddled picture concerning the issue of glutamatergic signaling [34]. Select studies have revealed site-specific glutamatergic dysfunction. Regions like the anterior cingulate cortex, prefrontal cortex (PFC), hippocampus, and occipital cortex have shown decreased glutamatergic signaling in depressed individuals [35–38]. Serum and CSF studies have been even less clear with studies showing both increased and decreased glutamate levels [28, 39, 40]. This dysfunction in glutamate has been attributed to various factors including HPA axis upregulation, elevated cytokines, and faulty glutamate recycling. Taken together, glutamate imbalance is a

part of the story of the pathophysiology of depression, but research has been showing that the glutamate story is more complicated than originally hypothesized.

The complexity and breadth of this topic have prompted many to call for an integration of parameters including neuroplasticity, synaptogenesis, and neuronal signal transduction. In essence, stress leads to weakening of the synapses within the rodent's brain leading to depression. As a result, it has been suggested that the "glutamate hypothesis" should be more broadly described as the "neuroplasticity hypothesis" [41, 42]. Neuroplasticity can be defined as the ability of the nervous system to adapt to stimuli, whether intrinsically or extrinsically [43]. Commonly, this is viewed as the brain's response to stress, and many diseases implicated with glutamatergic dysfunction are thought to be due to a maladaptive response to stress, otherwise referred to as poor or faulty neuroplasticity. Regardless, the past two decades have focused on uncovering the role of glutamate in depression. Studies have even shown that traditional antidepressants (e.g., SSRIs, SNRIs) work, in part, due to their effects on the glutamatergic system [44, 45]. Unfortunately, the precise glutamatergic effects of traditional antidepressants remain hazy in both basic science and clinical research. Since glutamatergic dysfunction in depression is now widely accepted, it is important to discuss one of the most implicated receptors, NMDAR, to further elaborate on this system.

## 21.6 The Role of NMDAR in MDD

As stated previously, NMDA receptors (NMDARs) are ligand-gated ion channels that are under the control of glutamate. There are four subunits that make up this receptor, and when activated by glutamate and its co-agonist glycine, there is an influx of cations. Most importantly, NMDAR allows the influx of $Ca^{2+}$ which can activate numerous signaling pathways within the neuron itself [23, 46]. It is important to note that for activation and opening of NMDAR, $Mg^{2+}$, a negative regulator of the channel, must be released. This occurs when an influx of cations enters the neuron, usually through AMPA or other glutamatergic receptors. As a result, NMDARs are typically activated only after other glutamatergic receptors on the neuron have been activated [23].

Once $Ca^{2+}$ flows into the cell, numerous reactions occur. The most important pathway for this discussion is the activation and synthesis of brain-derived neurotrophic factor (BDNF) [47–49]. Also known as abrineurin, BDNF, is a peptide growth factor that is essential for neuronal development, synaptogenesis, and neuroplasticity [50]. Low levels of BDNF have been implicated in depression and suicide in what is termed the "neurotrophin hypothesis" [51]. Dysfunction of glutamatergic signaling then can directly reduce BDNF levels, thus phenotypically presenting with depression or suicidal ideation. In the study of Murakami et al., both chronic and acute stress reduced BDNF mRNA expression in the hippocampus of rodents [52]. This link between stress and BDNF production supports the hypothesis that a dysfunctional stress response increases susceptibility to depression.

The ability to adapt to stress is in part controlled through glutamatergic signaling. NMDARs are critical for neuroplasticity through two mechanisms termed long-term potentiation (LTP) and long-term depression (LTD). These mechanisms allow glutamate to tightly modulate brain activity and appropriately respond to stress. In LTP, there is improved efficiency of certain connections, and in LTD, there is synaptic weakening. It has been suggested that through LTP and LTD, memory and cognition occur but also pathologic states such as depression [53]. The assumption here is that if certain synapses are altered due to dysfunction of NMDAR and glutamatergic signaling, depression or other pathologies can occur.

Dysfunction in NMDARs has been linked to depression, not only through changes in neuroplasticity but also in select studies examining receptor subunits. Specifically, Feyissa et al. showed that the NMDAR subunit NR2 is decreased in the prefrontal cortex (PFC) of postmortem brains of patients with MDD [54]. NR2 is a subunit critical to receptor function, and it is responsible for glutamate binding. Furthermore, postmortem studies have shown differences in the glutamate binding site of the NMDA receptor in the anterior PFC in suicide victims [55]. These studies indicate that there is NMDAR dysfunction in depressed patients, making it a clear target for therapeutics, such as ketamine and its derivative. In addition to NMDARs, there are other glutamatergic receptors that are critical to this topic, one of which is AMPAR.

## 21.7 The Role of AMPAR in MDD

AMPAR, or α-amino-3-hydroxyl-5-methyl-4-isoxazole-propionate receptor, is a four-subunit ionotropic receptor that is typically co-expressed with NMDAR on glutamatergic neurons [23]. When these receptors are activated by glutamate or AMPA, cations flow into the cell increasing its electric potential. This results in the activation of NMDAR allowing $Ca^{2+}$ influx and numerous downstream effects including BDNF production. Studies in rodents have shown that administration of ketamine leads to a rise in glutamate and activation of AMPAR [56–58]. Further, pre-treatment with an AMPAR antagonist, such as NBQX, reduces the antidepressive properties of ketamine indicating that AMPAR is critical to its efficacy [57]. Finally, postmortem studies in depressed patients have found reduced mRNA expression of AMPAR subunits in the brain, particularly in the hippocampus and cortex of the temporal lobe [22]. This highlights the role of AMPAR dysfunction in the pathophysiology of depression and how restoration of AMPAR function with ketamine leads to resolution of depression. Additionally, traditional antidepressants, such as fluoxetine and paroxetine, have been shown to upregulate AMPAR in rodents [59]. Through this data, one can see the clear implications of AMPAR in depression. Over the next sections, we will elaborate on the role of ketamine and esketamine in altering glutamatergic signaling via AMPAR and NMDAR to provide rapid-acting antidepressive effects.

## 21.8 History of Ketamine and Esketamine

Originally used as a veterinary anesthetic in the early 1960s, ketamine and its complex mechanism of action have been under investigation for over half a century. Its discovery occurred after intensive investigation of the anesthetic properties of phencyclidine (PCP) [9, 60]. Ketamine was found to be a powerful anesthetic in humans due to its ability to cause "dissociative anesthesia." This allows the patient to be awake yet have both amnesia and analgesia. These effects were first published in 1966 when Edward Domino and Guenter Corssen showed that ketamine could be used clinically to provide safe and effective anesthesia [61]. The relatively promising safety profile allowed the drug to be widely adopted as an anesthetic in the 1970s and still be used today. Unfortunately, ketamine became a popular recreational drug, and, as a result, restrictions surrounding its medical use became more stringent [60]. Ketamine is classified as an arylcyclohexylamine and is closely related structurally to PCP. Further, the racemic mixture contains both the S (+) and R (−) enantiomer.

As mentioned previously, ketamine was originally shown to have antidepressive effects in 2000, when Berman et al. published their influential data. These investigators demonstrated that a sub-anesthetic dose of 0.5 mg/kg exhibited improvement in depressive symptoms over 72 h in seven subjects [1]. This was dramatic because it introduced a potential novel therapy for treating depression and was unique due to its rapid acting effect. All of the other pharmacotherapies approved for depression act on monoamines and take 4–6 weeks to produce a noticeable effect. Berman's data caught the interest of those in psychiatric research leading to a significant amount of research published on its mechanism of action, pharmacodynamics, pharmacokinetics, and clinical effectiveness over the past two decades. The next few sections will highlight the available data, as they pertain to ketamine and its derivative.

## 21.9 Clinical Data Supporting Use in TRD

After Berman's pioneering study in 2000, it was not until 2006 that the same 0.5 mg/kg dose was shown by Zarate et al. to improve symptoms in a trial of 18 patients with TRD [62]. Following this trial, Mathew et al. showed that the use of ketamine was effective in a trial of 26 TRD patients in 2010 [63]. In this trial, 65% of patients responded to therapy at 24 h [63]. Numerous trials that followed between 2006 and 2017 further reaffirmed ketamine as a promising treatment in TRD, including reports by Ibrahim et al., Shiroma et al., and Murrough et al., among others [64–66]. Because of ketamine's clear benefit in TRD, many manufacturers were interested in developing the drug for depression treatment. One of those companies, Janssen Pharmaceuticals, sought to develop the S-enantiomer of ketamine, esketamine, in TRD and MDD with suicidality. One of the reasons that the S-enantiomer was chosen was due to the higher affinity to antagonize the NMDA receptor [67–69]. It has been stated that the S-enantiomer has four times greater potency for this receptor [67–69]. At the time this was believed to be beneficial, although there are now

mechanistic questions whether the S-enantiomer is preferred as an antidepressant [70, 71]. This discussion will be covered in the mechanism of action to follow. Janssen patented the intranasal formulation of the drug. This was helpful for dug development because up until that point, trials were conducted with the IV formulation.

In 2018, the phase II clinical trial in 67 TRD patients was published which showed that administration of esketamine in combination with traditional antidepressant drug therapy (i.e., SSRI, SNRI) leads to significant improvements in the Montgomery-Åsberg Depression Rating Scale (MADRS) in comparison with placebo [72]. Besides the significant efficacy with esketamine administration, there were numerous side effects that were noted in this trial. Several of these adverse events included transient elevations of blood pressure and heart rate, dissociative reactions, syncope, and headache [72]. These adverse events were not surprising based on the extensive history of investigation of this compound; however, they did bring to focus important practical considerations when administering this drug. Additionally, several treatment groups were left out of this study including those with a history of psychotic symptoms, bipolar disorder, alcohol or substance use disorders, recent use of marijuana, and current or recent suicidal ideation with intent to act [72]. Nevertheless, this trial was an important step in the approval of esketamine for patients with TRD.

Following this trial, numerous phase III clinical trials were published in 2019 and 2020 [73–76]. Not all of these trials showed significant therapeutic benefit; however, two of the trials showed significant positive results for esketamine and were used for submission to numerous regulatory bodies [73, 74]. These trials focused on the short-term benefit and time to relapse of esketamine in combination with oral antidepressants (AD). In the first study, there was a significant improvement of MADRS at 24 h when compared to placebo among 197 patients with TRD [74]. In the second study of 297 patients with TRD, the use of esketamine in combination with an oral AD was superior at delaying relapse in comparison with an oral AD alone [73]. However, following discontinuation of the active treatment trial, nearly 40% of patients in the esketamine group had relapsed by week 40 [73]. So, though esketamine delayed relapse, many patients eventually relapsed after the administration of esketamine was discontinued. Within these trials the adverse events were consistent from the phase II trial. Side effects included, but were not limited to, dissociation, somnolence, dizziness, and a rise in blood pressure [73, 74].

After the submission of this data to both the FDA and EMA, esketamine (Spravato) was approved in combination with an oral AD in March 2019 and December 2019, respectively [77]. Currently, there is only one other therapy, olanzapine-fluoxetine combination, approved for TRD. With the success of ketamine and esketamine in depression, there has been a significant amount of research into its mechanism of action. The following section will discuss the progress that has been made in uncovering the complex mechanism of ketamine and esketamine in depression.

## 21.10 Ketamine and Esketamine's Mechanism of Action: NMDAR and AMPAR

The mechanism of action of ketamine and its enantiomers has been under investigation since its original discovery in the 1960s. A few of the earliest papers on its mechanism of action were published in the 1980s, when it was found to reduce excitatory potentials in rodents, cats, and amphibians [78, 79]. This research was focused on the anesthetic properties of ketamine, and it was found that ketamine antagonized NMDA receptors on glutamatergic neurons. A simplified overview of the highly complex mechanism of action of these compounds is presented in Fig. 21.1. The way in which ketamine acts on NMDA is twofold. First, ketamine binds to a site within the channel of the NMDA receptor occluding cation flow. Additionally, ketamine acts by decreasing channel opening frequency through another allosteric mechanism [20, 80, 81]. As a result, inhibition of NMDA receptors on glutamatergic neurons leads to decreased activity in the CNS and results in dissociative anesthesia. These effects of ketamine were studied in the late twentieth century in depth.

The antidepressive properties of ketamine that occur when given at a sub-anesthetic dose were really investigated after Berman et al. published their results in 2000. As an aside, a sub-anesthetic dose is typically 0.1–0.5 mg/kg [82]. Studies have found that there are various cellular mechanisms of ketamine and its derivatives that result in its antidepressive effect [15, 83, 84]. The hypothesized mechanisms include (1) inhibition of NMDA receptors on GABAergic interneurons; (2) blockade of extra-synaptic NMDA receptors; and (3) activation of AMPAR resulting in mechanistic target of rapamycin complex (mTORC) activation and BDNF release.

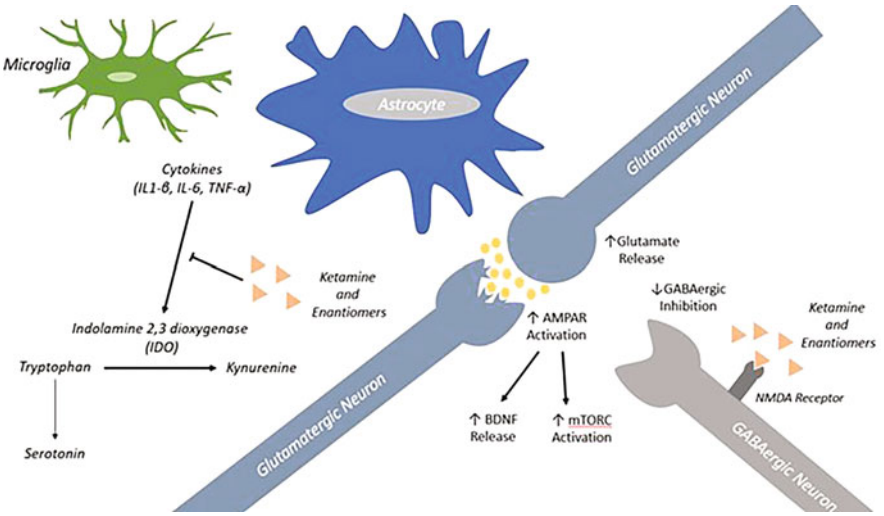


**Fig. 21.1** Simplified overview of the complex mechanism of action of ketamine and esketamine

These are just a few of the mechanisms that have been attributed to the antidepressive effects. Outside of NMDA, ketamine and its enantiomers have affinity for serotonin reuptake transporters (SERT), norepinephrine reuptake transporters (NET), sigma-1 receptors, and opioid receptors [84, 85]. Ketamine also has been shown to reduce activation of the lateral habenula which is referred to as the "anti-reward center." It is postulated that by downregulating the lateral habenula, ketamine is further able to improve depression [86]. It is important to acknowledge, however, that as more data emerge, many neuroscientists are questioning whether NMDA antagonism is truly the primary mechanism behind the antidepressive efficacy of ketamine and esketamine [84].

With that said, one of the mechanisms that has been extensively investigated is the inhibition of NMDA receptors on GABAergic interneurons. Ketamine decreases inhibition from the GABAergic interneurons, thereby increasing glutamatergic signaling. This effect of ketamine and esketamine results in activation of neurons, particularly pyramidal neurons, in the cerebral cortex [81, 84, 87]. As a result, there is an increase in CNS activity and associated decrease in depressive symptoms. Rodent studies have shown an increase in glutamate concentration after the administration of ketamine [88].

Additionally, there is inhibition of extrasynaptic NMDA receptors by ketamine and its derivatives. These extrasynaptic receptors are located on cortical neurons and act to suppress cortical neuronal function. These receptors are activated by basal glutamate levels in the brain. After administration of ketamine, inhibition of these receptors frees up cortical pyramidal neurons and results in their activation [81, 84]. This has been shown in mouse models as an additional mechanism by which ketamine reduces depression [89].

One mechanism that has seen growing support as the primary mechanism for ketamine's effectiveness in depression is the activation of AMPAR. This activation occurs because of the increased glutamate that is released as a result of GABAergic interneuron blockade. AMPAR signaling results in numerous downstream effects. One these effects is activation of mTORC, a critical pathway in synaptogenesis [58, 84, 90, 91]. This mechanism is of particular importance based on rodent studies showing that infusion of rapamycin, an inhibitor of mTORC, resulted in decreased antidepressive effects of ketamine [92]. With that said, studies in humans showed that rapamycin administration was unable to attenuate the antidepressive effects of ketamine and increased the response rates at 2 weeks [93]. In one rodent study, inhibition of mTORC blocked the effects of esketamine, but not R-ketamine [69]. This highlights the intricate biological mechanism with mTORC and the importance of distinguishing enantiomers.

Besides mTORC, AMPAR activation has been shown to result in BDNF release [13, 58, 81]. BDNF, a potent neurotrophic factor, is hypothesized to be critical in the rapid acting effect of ketamine and its enantiomers. One mechanism by which this occurs is through inhibition of eukaryotic elongation factor 2 (eEF2), an inhibitor of BDNF translation [94]. Activation of AMPAR via a glutamatergic surge results in a positive feedback loop and additional BDNF translation via tropomyosin receptor kinase B (TrkB) signaling [15, 94]. This loop releases a large amount of BDNF and

is one of the many distinguishing factors between ketamine, esketamine, and traditional oral ADs. While oral ADs result in increased expression of BDNF over several weeks, there is a decreased initial release of BDNF with traditional ADs in comparison with ketamine and esketamine [15].

While numerous studies have implicated a critical role of NMDA antagonism in ketamine's efficacy, it is hypothesized that there are additional mechanisms that are not yet known. Studies with other NMDA antagonists, such as memantine, a treatment used in Alzheimer's disease, have not shown any antidepressive effects [95]. Therefore, it is hypothesized that other serotonergic, dopaminergic, noradrenergic, or opioid signaling may play a role, and it is unlikely that NMDA antagonism alone is driving the antidepressive effects [84]. Furthermore, the degradation products of ketamine and its enantiomers are drivers for its efficacy. Particularly, the production of (S,R)-hydroxynorketamine (S,R-HNK) may also contribute to its antidepressive effectiveness [96, 97].

Rodent studies have shown mixed results with S,R-HNK [68, 96, 98]. As a result, the role that HNK plays in ketamine and esketamine's mechanism of action is still uncertain. Select rodent studies have shown that the metabolite S-norketamine produced a response similar to esketamine [97]. Further, S-norketamine did not show many of the dissociative effects of esketamine [97]. In summary, additional studies in humans are necessary to better understand these complex actions of ketamine and esketamine.

Finally, it is important to distinguish between mechanisms associated with S-ketamine (i.e., esketamine) and R-ketamine. Esketamine has been shown to be a more potent inhibitor of the NMDA receptor [67]. Nevertheless, there has been increasing literature that supports the clinical use of R-ketamine. The reasons for this are largely related to a proposed similar efficacy and reduced dissociation induced by R-ketamine [70, 71, 97]. In Yang et al.'s study in rodents, administration of R-ketamine resulted in a more potent response and longer-lasting antidepressant effects than S-ketamine [70]. Further, previous studies have shown that S-ketamine elicits greater undesirable psychotomimetic side effects (e.g., dissociation, hallucinations) than R-ketamine [70, 99]. Unfortunately, there is limited published data directly comparing S-ketamine and R-ketamine on TRD in humans. Such a study would be helpful to not only determine which is the more effective and safe treatment but also help understand the role NMDA antagonism truly plays. If R-ketamine, the less potent NMDA antagonist, is shown to have greater antidepressive properties, then it can be deduced that NMDA antagonism plays a less prominent role in the mechanism of action.

A body of data has been published evaluating racemic ketamine vs. esketamine. These studies have found that IV racemic ketamine exhibits a greater overall response and remission rates in comparison with intranasal esketamine [100]. Unfortunately, from years of use as an anesthetic, racemic ketamine is generic, and manufacturers are unlikely to complete clinical trials necessary for approval in depression.

In conclusion, ketamine and esketamine have a complex mechanism of action that has been investigated at length. While there is increased glutamatergic signaling,

additional studies are necessary to confirm many hypotheses regarding the mechanism(s) behind it. One area to watch is further studies on R-ketamine. With less potent NMDA antagonism, it is surprising that preclinical rodent studies have shown increased antidepressive response and decreased dissociation with its administration [70, 71, 101]. As we will discuss in the following section, the dissociative adverse effects are one of the many drawbacks of esketamine therapy that prevents its more widespread use. The next section will review the benefits and drawbacks of esketamine therapy in TRD.

## 21.11 Benefits and Drawbacks of Therapy

Intranasal esketamine (Spravato) offers several benefits to patients with TRD. These benefits include (1) a rapid onset of effect; (2) promising efficacy when few other treatment options exist; and (3) a potent anti-suicidality effect.

The first and probably the most significant is the rapid acting antidepressive effect. Patients who receive esketamine reported efficacy almost immediately after administration with increasing antidepressive effect upon repeat dosing. In one of the phase III trials, there was improvement of >3 points in MADRS in the esketamine and oral antidepressant (AD) group vs. placebo and oral AD within the first 24 h [74]. By day 28, this difference between the two groups increased to 4 points, and the total change in the treatment group was an improvement of 21.4 points [74]. The clinical data clearly show the rapid acting effect of adjunctive esketamine over traditional oral AD alone. As stated previously, the rationale behind this rapid acting effect is still unclear. One hypothesis is that esketamine results in the release of BDNF, whereas traditional oral AD results in greater expression of BDNF over many weeks [15].

Outside of the rapid acting effect, another benefit of esketamine is its efficacy in a disease area with very few treatments. As stated previously, olanzapine-fluoxetine combination is the only other pharmacotherapy approved for TRD. As a result, the approval of esketamine offered new hope for patients with TRD.

Finally, esketamine has a potent anti-suicidality effect that allows it to be extremely valuable in emergency situations of patients with major depression with suicidal ideation (MDSI). This indication was approved in the United States in August 2020 for MDSI and became the first therapy approved with rapid symptom control in MDD. There were two phase II clinical trials, ASPIRE I and ASPIRE II, which showed statistically significant improvement in depressive symptoms at 24 h [102, 103].

Despite all the benefits, there are some drawbacks with esketamine treatment. A few of these include (1) adverse effects such as dissociation and transient rise in blood pressure; (2) a demanding treatment regimen; and (3) limited durability of response.

One of the drawbacks of esketamine treatment is the adverse event profile. Esketamine has been approved with a black box label, the most severe safety warnings given by the FDA [77]. This is due to the risk of sedation, dissociation,

abuse and misuse, and suicidal thoughts in select patients. Besides these potentially severe side effects, esketamine is also associated with dizziness, nausea, hypoesthesia, anxiety, lethargy, vomiting, and rise in blood pressure [77]. These side effects occurred at an incidence of at least 5% in clinical studies, but for the most part they are transient and resolve within a few hours post-administration [77].

Because of the risk of adverse events, esketamine has additional barriers to treatment that have been mandated by the FDA. For instance, esketamine must be administered at a qualified health facility by an esketamine certified prescriber. Additionally, patients must be observed for 2 h following administration, and the patient is unable to drive for 24 h. These barriers alone can be very restrictive, if the patient has a job or has difficulty getting to a certified health center. This is especially true due to the dosing regimen. With esketamine, the induction period consists of twice weekly treatments for 4 weeks followed by once weekly treatments for weeks 5–8 [77]. After week 8, treatments are recommended weekly or biweekly [77]. This is a significant commitment and is not suitable for all patients. As a result, when prescribing this therapy, it is imperative that the prescriber discusses potential risks and requirements.

A final drawback to consider is the limited durability of response in many patients. In clinical trials, nearly 40% of patients experienced a relapse of their depression by week 40 while on esketamine and an oral AD [73]. This was improved in comparison with 50% of patients on AD and placebo. Nevertheless, there is still significant room for improvement particularly when it comes to long-term treatment for TRD. Based on our own experience to date, we believe booster treatments at specified intervals will assure long-term maintenance of the beneficial outcome. However, precise recommendations about booster efficacy and administration schedules will have to await longer-term clinical trials.

While this description of the benefits and drawbacks is not exhaustive, it does highlight many important considerations. The following section will discuss another perspective in the treatment of depression and especially TRD and the mechanism behind ketamine, that is, the role of inflammation.

## 21.12 Role of Inflammation in MDD and TRD

Substantial evidence links inflammatory pathways to changes in glutamatergic and monoaminergic signaling resulting in clinical depression [29, 104, 105]. This has been reported in numerous studies and led to the development of the field psychoneuroimmunology [106–108]. Pioneered by studies completed by Ader and Cohen nearly 50 years ago, this field has grown into a massive area of psychiatric research [106]. While many books have been published on this subject, this section will give an overview of the field with a particular focus on the impact of glutamate. It has been consistently shown that inflammation is increased in patients with MDD and TRD [109–114]. This occurs through numerous pathways that have been covered at length. The focus of this chapter, however, will be on: (1) activation of indoleamine 2,3-dioxygenase; (2) cytokine-induced glutamatergic excitotoxicity; and

(3) stress-induced inhibition of glutamatergic signaling. It should first be restated, however, that the glutamatergic system is one of the most tightly regulated systems within the brain. Both an excess of and a deficiency in glutamate can lead to severe psychiatric conditions, such as depression. This occurs through various mechanisms, and this section will highlight a few as it relates to inflammation.

One of the most extensively investigated areas of research surrounding inflammation and depression involves indoleamine 2,3-dioxygenase (IDO) [115–117]. This enzyme is found in the intestines, lungs, female genital tract, placenta, and lymphatic tissue, such as microglial cells. It is a critical mediator in the breakdown of tryptophan in the kynurenine pathway which ultimately leads to the production of nicotinamide adenine dinucleotide. While this pathway is critical in cell metabolism, if upregulated, it can divert tryptophan away from the serotonin pathway where it is also a precursor [116, 118]. In conjunction with inflammation, this pathway can be upregulated through various signaling cascades (e.g., MAPK, NFkB). Additionally, high levels of kynurenine can lead to cell-specific conversion into other toxic products within the brain. For instance, within astrocytes, kynurenine can be converted to kynurenic acid (KA). High levels of KA are believed to result in inhibition of glutamatergic signaling [104, 118]. As a result, there can be decreased dopamine release within the striatum [104, 118]. This has been shown in rodent models [119]. On the other hand, within glial cells, kynurenine is converted to quinolinic acid (QA), a toxic metabolite [120]. QA has been shown to activate NMDA receptors leading to additional glutamate release [121]. With excessive glutamate, excitotoxicity can occur, and BDNF production is decreased. Although the exact mechanism of neurotoxicity is still being investigated, studies have shown that excess QA leads to decreased BDNF in rodents [122]. As discussed previously, decreased levels of BDNF have been linked to depression and suicidality [15, 49]. These are two ways by which inflammation can lead to depressive symptoms through both decreased glutamatergic and dopaminergic signaling in the striatum, as well as glutamate excitotoxicity and decreased BDNF production in the PFC and hippocampus. Admittedly, much of this research is preclinical, and clinical studies are necessary to confirm this mechanism.

In addition to the impact of inflammation and IDO, there are also direct implications of cytokine release on glutamatergic signaling. Increased cytokines (e.g., TNF-alpha, IL-1, IL-2, IL-6) and acute phase reactants (e.g., C-reactive protein) have been directly linked to depression for years [112, 123–126]. This has been a target of therapeutics through utilizing anti-inflammatories, such as celecoxib, to enhance depression treatment [127–130]. Studies have also shown that depression is a common side effect in those utilizing cytokines, such as IFN-alpha, for cancer or hepatitis C [131–133]. Several mechanisms have been proposed as to why cytokine release may result in depression, such as HPA upregulation, IDO activation, and serotonergic and noradrenergic dysregulation. One mechanism that is particularly relevant to this chapter is the impact cytokines have on glutamate. It has been shown that cytokines dramatically impact glutamate recycling [104, 112, 134]. This occurs when cytokines increase glutamate release and decrease glutamate reuptake in astrocytes and other glial cells [104, 112, 134]. The increase of glutamate from

astrocytes leads to a preferential binding to extra-synaptic NMDA receptors. As stated in the mechanisms of action section above, these receptors are tonic regulators of cortical function and BDNF production. Increased glutamate binding through cytokine activation leads to decreased cortical activity and BDNF production, further contributing to depression.

Finally, the close link between inflammation, stress, and glutamate excitotoxicity may also contribute to depression. For example, corticotrophin-releasing hormone (CRH) is released by the hypothalamus and is a critical component of the stress response within the HPA axis [135, 136]. As a result of this increased secretion, there is increased HPA activity. Studies have shown HPA upregulation to be linked to depression and other mood disorders [137, 138]. In terms of glutamatergic signaling, it has been shown that chronic stress induces changes in this system in two ways. First, HPA upregulation leads to increased glutamatergic signaling and excitotoxicity. This excitotoxic surge of glutamate is followed by adaptive downregulation of glutamatergic activation leading to depression-like symptoms in rodents [52, 112, 139]. This is clearly detectable in the PFC and hippocampus in rodent studies [112, 139]. Indeed, in select rodent studies, HPA upregulation and glutamatergic excitotoxicity were shown to lead to volume reduction in the PFC and hippocampus [140]. As a result, research has confirmed the stress response impacts the glutamatergic system and may play a role in the pathophysiology of depression. Unfortunately, limited progress has been made in considering the hyperactive HPA axis therapeutically.

Overall, the field of psychoneuroimmunology is an exciting area of scientific endeavor, and our understanding of glutamate's role within and as a result of inflammation is expanding. Activation of IDO, increased cytokine release, and HPA axis upregulation alter the glutamatergic system and have downstream effects manifesting themselves as depression. Outside of traditional anti-inflammatory drugs, such as NSAIDs, limited pharmacotherapeutics are available to treat psychiatric conditions through principally targeting inflammation. With growing research and clinical use, ketamine and its enantiomers have been shown to impact inflammation in a way unlike other currently marketed antidepressants.

## 21.13 Ketamine, Esketamine, and Inflammation

Over the past 50 years, a significant debate has occurred regarding the role of ketamine and its enantiomers in reducing inflammation. In fact, some authors argue that it is not an anti-inflammatory agent, but rather an anti-pro-inflammatory agent [141]. That is, ketamine reduces the pro-inflammatory response. This has been under investigation since ketamine was shown to be beneficial as an anesthetic agent in septic shock. Studies have largely rationalized its benefit in septic shock to be principally due to its preservation of cardiovascular function compared with other anesthetics [142–144]. Outside of the cardiovascular benefit of ketamine, there is also an anti-inflammatory benefit of utilizing ketamine due to its inhibition of cytokine release, particularly TNF-alpha and IL-6 [145, 146]. This has been

investigated in numerous studies with ketamine as an anesthetic and more recently as an antidepressant [144–147]. As a result, there is both decreased activation of IDO and decreased HPA hyperactivation. This is an important link to ways in which ketamine may rapidly decrease depression in patients who have been treatment resistant. Studies have indicated that levels of inflammation are correlated with treatment resistance in major depression [148]. In fact, it may be the anti-inflammatory properties of ketamine that are making a difference in the treatment-resistant population.

Decreased activation of IDO due to decreased cytokine production has been reported in numerous human and animal studies. This result was established both by decreased IDO activation and decreased KYN/tryptophan ratio [141, 149–151]. As stated above, upregulation of IDO has been identified as a pathophysiological mechanism of depression [115]. These studies indicate that ketamine's complex mechanism likely reduces activation of the kynurenine pathway and thus reduces tryptophan depletion, an important precursor for serotonin.

Additionally, ketamine is also believed to normalize the HPA axis. Through decreased release of inflammatory cytokines and acute phase reactants, ketamine is able to decrease the activation of the HPA axis in numerous rodent studies [151, 152]. In one study, the authors suggest that chronic stress on rodents led to HPA hyperactivation and depressive-like behavior in part due to downregulation of the glucocorticoid receptor within the hippocampus and upregulation of corticosterone, the primary stress hormone in rodents [151]. Upon administration of ketamine, the rodents resumed normal behavior and were shown to have return of baseline levels of the glucocorticoid receptor in the hippocampus as well as normalized corticosterone levels [151]. Additional studies in humans monitoring HPA axis activity are necessary as much of the data to date are unclear.

As stated above, while many studies in both humans and animals predict an anti-inflammatory effect of ketamine, it is unclear if this effect is the cause of its efficacy or simply just another ancillary effect. Certain studies point to downward trends in serum cytokines to be associated with treatment response to ketamine and depression relief [153]. On the other hand, select studies found only transient decreases in cytokines bringing to question the role inflammation plays in ketamine's effectiveness [146, 154]. One thing that is clear, however, is the need for more clinical investigation into this matter.

## 21.14 Future Direction

The approval of esketamine arrived after years of investigation and brought new hope into the treatment of TRD. This agent was the first to be approved for the treatment of TRD since 2009 and a novel mechanism targeting the glutamatergic system. As we look ahead, there are over 100 clinical trials in the United States currently underway in TRD [155]. These trials are investigating agents such as novel small molecules, nerve stimulators, and older known compounds such as psilocybin [155]. Additionally, there are numerous investigators studying the glutamatergic

system and its role in depression, ketamine's complex mechanism, and other pieces to this puzzle. As we look ahead, there are several important questions that will need to be answered. How does dysregulation of the glutamatergic system first occur to cause depression? Is there a role for R-ketamine in TRD? Does inflammation cause depression, or is it merely associated with the disease? How can duration of response be improved with esketamine in TRD? Significant progress has been made to answer these questions over the past few decades. As we look toward the future, additional information will provide new avenues to target TRD.

## 21.15 Conclusion

In summary, this chapter defined treatment-resistant depression and discussed current understanding of prevalence, pathophysiology, and treatments with an emphasis on the glutamatergic system. As described previously, the glutamatergic system is tightly regulated, and imbalance within it can lead to various pathologies. It is an extremely intricate system that is critical to cognition, learning, and memory and more recently understood to play a role in mood. Additionally, we highlighted the history and current understanding of the only glutamatergic agent approved for the treatment of TRD, esketamine. This agent can normalize glutamatergic activity in various areas of the brain and increase important neurotropic factors such as BDNF. We discussed the current understanding of inflammation in depression and how ketamine has been shown to influence it. This includes ketamine's role on the kynurenine pathway and HPA axis principally through its reduction of inflammatory mediators. With this in mind, we briefly discussed the future direction of the treatment of TRD and questions that remain unanswered. Ultimately, the information presented in this chapter can be used to guide research and improve the treatment for this debilitating disease.

## References

1. Berman RM, Cappiello A, Anand A, Oren DA, Heninger GR, Charney DS, et al. Antidepressant effects of ketamine in depressed patients. Biol Psychiatry. 2000;47(4):351–4.
2. Crupi R, Marino A, Cuzzocrea S. New therapeutic strategy for mood disorders. Curr Med Chem. 2011;18(28):4284–98.
3. Rosenblat JD, McIntyre RS, Alves GS, Fountoulakis KN, Carvalho AF. Beyond monoamines-novel targets for treatment-resistant depression: a comprehensive review. Curr Neuropharmacol. 2015;13(5):636–55.
4. World Health Organization. Depression fact sheet [Internet]. 2020. https://www.who.int/news-room/fact-sheets/detail/depression. Accessed 17 Sep 2021.
5. Center for Disease Control and Prevention. Suicide and self-inflicted injury. 2020. https://www.cdc.gov/nchs/fastats/suicide.htm. Accessed 18 Sep 2021.
6. Institute of Health Metrics and Evaluation. Health data exchange. 2021. http://ghdx.healthdata.org/gbd-results-tool?params=gbd-api-2019-permalink/d780dffbe8a381b25e1416884959e88b. Accessed 7 Sep 2021.

7. Bunney WE Jr, Davis JM. Norepinephrine in depressive reactions. A review. Arch Gen Psychiatry. 1965;13(6):483–94.
8. Delgado PL, Moreno FA. Role of norepinephrine in depression. J Clin Psychiatry. 2000;61(1): 5–12.
9. Hillhouse TM, Porter JH. A brief history of the development of antidepressant drugs: from monoamines to glutamate. Exp Clin Psychopharmacol. 2015;23(1):1–21.
10. Cole CE, Patterson RM, Craig JB, Thomas WE, Ristine LP, Stahly M, et al. A controlled study of efficacy of iproniazid in treatment of depression. AMA Arch of Gen Psychiatry. 1959;1(5): 513–8.
11. Fisar Z, Hroudová J, Raboch J. Inhibition of monoamine oxidase activity by antidepressants and mood stabilizers. Neuro Endocrinol Lett. 2010;31(5):645–56.
12. Gonul AS, Akdeniz F, Taneli F, Donat O, Eker C, Vahip S. Effect of treatment on serum brain-derived neurotrophic factor levels in depressed patients. Eur Arch Psychiatry Clin Neurosci. 2005;255(6):381–6.
13. Machado-Vieira R, Yuan P, Brutsche N, DiazGranados N, Luckenbaugh D, Manji HK, et al. Brain-derived neurotrophic factor and initial antidepressant response to an N-methyl-D-aspartate antagonist. J Clin Psychiatry. 2009;70(12):1662–6.
14. Homberg JR, Molteni R, Calabrese F, Riva MA. The serotonin-BDNF duo: developmental implications for the vulnerability to psychopathology. Neurosci Biobehav Rev. 2014;43:35–47.
15. Björkholm C, Monteggia LM. BDNF—a key transducer of antidepressant effects. Neuropharmacology. 2016;102:72–9.
16. Buetler L, Clarkin J, Bongar B. Guideline for the systematic treatment of the depressed patient. Oxford Scholarship Online. Oxford: Oxford University Press; 2000.
17. McIntyre RS, Filteau MJ, Martin L, Patry S, Carvalho A, Cha DS, et al. Treatment-resistant depression: definitions, review of the evidence, and algorithmic approach. J Affect Disord. 2014;156:1–7.
18. Gaynes B, Asher G, Gartlehner G, Hoffman V, Cokker-Schwimmer E. Definition of treatment-resistant depression in the medicare population. Rockville, MD: Agency for Healthcare Research and Quality (US); 2018.
19. Fonnum F. Glutamate: a neurotransmitter in mammalian brain. J Neurochem. 1984;42(1): 1–11.
20. Trullas R, Skolnick P. Functional antagonists at the NMDA receptor complex exhibit antidepressant actions. Eur J Pharmacol. 1990;185(1):1–10.
21. Curtis DR, Watkins JC. The excitation and depression of spinal neurones by structurally related amino acids. J Neurochem. 1960;6:117–41.
22. Beneyto M, Kristiansen LV, Oni-Orisan A, McCullumsmith RE, Meador-Woodruff JH. Abnormal glutamate receptor expression in the medial temporal lobe in schizophrenia and mood disorders. Neuropsychopharmacology. 2007;32(9):1888–902.
23. Traynelis SF, Wollmuth LP, McBain CJ, Menniti FS, Vance KM, Ogden KK, et al. Glutamate receptor ion channels: structure, regulation, and function. Pharmacol Rev. 2010;62(3):405–96.
24. Zarate C Jr, Machado-Vieira R, Henter I, Ibrahim L, Diazgranados N, Salvadore G. Glutamatergic modulators: the future of treating mood disorders? Harv Rev Psychiatry. 2010;18(5):293–303.
25. Drago A, Crisafulli C, Sidoti A, Serretti A. The molecular interaction between the glutamatergic, noradrenergic, dopaminergic and serotoninergic systems informs a detailed genetic perspective on depressive phenotypes. Prog Neurobiol. 2011;94(4):418–60.
26. Andersen JV, Markussen KH, Jakobsen E, Schousboe A, Waagepetersen HS, Rosenberg PA, et al. Glutamate metabolism and recycling at the excitatory synapse in health and neurodegeneration. Neuropharmacology. 2021;196:108719.
27. Hynd MR, Scott HL, Dodd PR. Glutamate-mediated excitotoxicity and neurodegeneration in Alzheimer's disease. Neurochem Int. 2004;45(5):583–95.

28. Kim JS, Schmid-Burgk W, Claus D, Kornhuber HH. Increased serum glutamate in depressed patients. Arch Psychiatr Nervenkr. 1982;232(4):299–304.
29. Tilleux S, Hermans E. Neuroinflammation and regulation of glial glutamate uptake in neurological disorders. J Neurosci Res. 2007;85(10):2059–70.
30. Iovino L, Tremblay ME, Civiero L. Glutamate-induced excitotoxicity in Parkinson's disease: the role of glial cells. J Pharmacol Sci. 2020;144(3):151–64.
31. Li CT, Yang KC, Lin WC. Glutamatergic dysfunction and glutamatergic compounds for major psychiatric disorders: evidence from clinical neuroimaging studies. Front Psych. 2019;9:767.
32. Niswender CM, Conn PJ. Metabotropic glutamate receptors: physiology, pharmacology, and disease. Annu Rev Pharmacol Toxicol. 2010;50:295–322.
33. Crane G. Cyloserine as an antidepressant agent. Am J Psychiatry. 1959;115(11):1025–6.
34. Yüksel C, Öngür D. Magnetic resonance spectroscopy studies of glutamate-related abnormalities in mood disorders. Biol Psychiatry. 2010;68(9):785–94.
35. Auer DP, Pütz B, Kraft E, Lipinski B, Schill J, Holsboer F. Reduced glutamate in the anterior cingulate cortex in depression: an in vivo proton magnetic resonance spectroscopy study. Biol Psychiatry. 2000;47(4):305–13.
36. Hasler G, van der Veen JW, Tumonis T, Meyers N, Shen J, Drevets WC. Reduced prefrontal glutamate/glutamine and gamma-aminobutyric acid levels in major depression determined using proton magnetic resonance spectroscopy. Arch Gen Psychiatry. 2007;64(2):193–200.
37. Block W, Träber F, von Widdern O, Metten M, Schild H, Maier W, et al. Proton MR spectroscopy of the hippocampus at 3 T in patients with unipolar major depressive disorder: correlates and predictors of treatment response. Int J Neuropsychopharmacol. 2009;12(3):415–22.
38. Clark DL, MacMaster FP, Brown EC, Kiss ZHT, Ramasubbu R. Rostral anterior cingulate glutamate predicts response to subcallosal deep brain stimulation for resistant depression. J Affect Disord. 2020;266:90–4.
39. Frye MA, Tsai GE, Huggins T, Coyle JT, Post RM. Low cerebrospinal fluid glutamate and glycine in refractory affective disorder. Biol Psychiatry. 2007;61(2):162–6.
40. Moriguchi S, Takamiya A, Noda Y, Horita N, Wada M, Tsugawa S, et al. Glutamatergic neurometabolite levels in major depressive disorder: a systematic review and meta-analysis of proton magnetic resonance spectroscopy studies. Mol Psychiatry. 2019;24(7):952–64.
41. Pittenger C, Duman RS. Stress, depression, and neuroplasticity: a convergence of mechanisms. Neuropsychopharmacology. 2008;33(1):88–109.
42. Sanacora G, Treccani G, Popoli M. Towards a glutamate hypothesis of depression: an emerging frontier of neuropsychopharmacology for mood disorders. Neuropharmacology. 2012;62(1):63–77.
43. Mateos-Aparicio P, Rodríguez-Moreno A. The impact of studying brain plasticity. Front Cell Neurosci. 2019;13:66.
44. Barbon A, Popoli M, La Via L, Moraschi S, Vallini I, Tardito D, et al. Regulation of editing and expression of glutamate alpha-amino-propionic-acid (AMPA)/kainate receptors by antidepressant drugs. Biol Psychiatry. 2006;59(8):713–20.
45. Bleakman D, Alt A, Witkin JM. AMPA receptors in the therapeutic management of depression. CNS Neurol Disord Drug Targets. 2007;6(2):117–26.
46. Schoepfer R, Monyer H, Sommer B, Wisden W, Sprengel R, Kuner T, et al. Molecular biology of glutamate receptors. Prog Neurobiol. 1994;42(2):353–7.
47. Du J, Feng L, Yang F, Lu B. Activity- and Ca(2+)-dependent modulation of surface expression of brain-derived neurotrophic factor receptors in hippocampal neurons. J Cell Biol. 2000;150(6):1423–34.
48. Lee B, Kim Y. The roles of BDNF in the pathophysiology of major depression and in antidepressant treatment. Psychiatry Investig. 2010;7(4):231–5.
49. Yu H, Chen ZY. The role of BDNF in depression on the basis of its location in the neural circuitry. Acta Pharmacol Sin. 2011;32(1):3–11.

50. Kowiański P, Lietzau G, Czuba E, Waśkow M, Steliga A, Moryś J. BDNF: a key factor with multipotent impact on brain signaling and synaptic plasticity. Cell Mol Neurobiol. 2018;38(3):579–93.
51. Dwivedi Y. Brain-derived neurotrophic factor: role in depression and suicide. Neuropsychiatr Dis Treat. 2009;5:433–49.
52. Murakami S, Imbe H, Morikawa Y, Kubo C, Senba E. Chronic stress, as well as acute stress, reduces BDNF mRNA expression in the rat hippocampus but less robustly. Neurosci Res. 2005;53(2):129–39.
53. Bliss TV, Cooke SF. Long-term potentiation and long-term depression: a clinical perspective. Clinics (Sao Paulo). 2011;66(1):3–17.
54. Feyissa AM, Chandran A, Stockmeier CA, Karolewicz B. Reduced levels of NR2A and NR2B subunits of NMDA receptor and PSD-95 in the prefrontal cortex in major depression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2009;33(1):70–5.
55. Nowak G, Ordway GA, Paul IA. Alterations in the N-methyl-D-aspartate (NMDA) receptor complex in the frontal cortex of suicide victims. Brain Res. 1995;675(1–2):157–64.
56. Martinez-Turrillas R, Frechilla D, Del Río J. Chronic antidepressant treatment increases the membrane expression of AMPA receptors in rat hippocampus. Neuropharmacology. 2002;43(8):1230–7.
57. Koike H, Iijima M, Chaki S. Involvement of AMPA receptor in both the rapid and sustained antidepressant-like effects of ketamine in animal models of depression. Behav Brain Res. 2011;224(1):107–11.
58. Zhou W, Wang N, Yang C, Li XM, Zhou ZQ, Yang JJ. Ketamine-induced antidepressant effects are associated with AMPA receptors-mediated upregulation of mTOR and BDNF in rat hippocampus and prefrontal cortex. Eur Psychiatry. 2014;29(7):419–23.
59. Svenningsson P, Tzavara ET, Witkin JM, Fienberg AA, Nomikos GG, Greengard P. Involvement of striatal and extrastriatal DARPP-32 in biochemical and behavioral effects of fluoxetine (Prozac). Proc Natl Acad Sci. 2002;99(5):3182–7.
60. Liao Y, Tang YL, Hao W. Ketamine and international regulations. Am J Drug Alcohol Abuse. 2017;43(5):495–504.
61. Corssen G, Domino EF. Dissociative anesthesia: further pharmacologic studies and first clinical experience with the phencyclidine derivative CI-581. Anesth Analg. 1966;45(1):29–40.
62. Zarate CA Jr, Singh JB, Carlson PJ, Brutsche NE, Ameli R, Luckenbaugh DA, et al. A randomized trial of an N-methyl-D-aspartate antagonist in treatment-resistant major depression. Arch Gen Psychiatry. 2006;63(8):856–64.
63. Mathew SJ, Murrough JW, Aan Het Rot M, Collins KA, Reich DL, Charney DS. Riluzole for relapse prevention following intravenous ketamine in treatment-resistant depression: a pilot randomized, placebo-controlled continuation trial. Int J Neuropsychopharmacol. 2010;13(1):71–82.
64. Ibrahim L, Diazgranados N, Luckenbaugh DA, Machado-Vieira R, Baumann J, Mallinger AG, et al. Rapid decrease in depressive symptoms with an N-methyl-d-aspartate antagonist in ECT-resistant major depression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2011;35(4):1155–9.
65. Murrough JW, Iosifescu DV, Chang LC, Al Jurdi RK, Green CE, Perez AM, et al. Antidepressant efficacy of ketamine in treatment-resistant major depression: a two-site randomized controlled trial. Am J Psychiatry. 2013;170(10):1134–42.
66. Shiroma PR, Johns B, Kuskowski M, Wels J, Thuras P, Albott CS, et al. Augmentation of response and remission to serial intravenous subanesthetic ketamine in treatment resistant depression. J Affect Disord. 2014;155:123–9.
67. Domino EF. Taming the ketamine tiger—1965. Anesthesiology. 2010;113(3):678–84.
68. Zanos P, Moaddel R, Morris PJ, Georgiou P, Fischell J, Elmer GI, et al. NMDAR inhibition-independent antidepressant actions of ketamine metabolites. Nature. 2016;533(7604):481–6.

69. Yang C, Ren Q, Qu Y, Zhang JC, Ma M, Dong C, et al. Mechanistic target of rapamycin-independent antidepressant effects of (R)-ketamine in a social defeat stress model. Biol Psychiatry. 2018;83(1):18–28.
70. Yang C, Shirayama Y, Zhang JC, Ren Q, Yao W, Ma M, et al. R-ketamine: a rapid-onset and sustained antidepressant without psychotomimetic side effects. Transl Psychiatry. 2015;5(9):e632.
71. Zhang JC, Li SX, Hashimoto K. R (−)-ketamine shows greater potency and longer lasting antidepressant effects than S (+)-ketamine. Pharmacol Biochem Behav. 2014;116:137–41.
72. Daly EJ, Singh JB, Fedgchin M, Cooper K, Lim P, Shelton RC, et al. Efficacy and safety of intranasal esketamine adjunctive to oral antidepressant therapy in treatment-resistant depression: a randomized clinical trial. JAMA Psychiat. 2018;75(2):139–48.
73. Daly EJ, Trivedi MH, Janik A, Li H, Zhang Y, Li X, et al. Efficacy of esketamine nasal spray plus oral antidepressant treatment for relapse prevention in patients with treatment-resistant depression: a randomized clinical trial. JAMA Psychiat. 2019;76(9):893–903.
74. Popova V, Daly EJ, Trivedi M, Cooper K, Lane R, Lim P, et al. Efficacy and safety of flexibly dosed esketamine nasal spray combined with a newly initiated oral antidepressant in treatment-resistant depression: a randomized double-blind active-controlled study. Am J Psychiatry. 2019;176(6):428–38.
75. Fedgchin M, Trivedi M, Daly EJ, Melkote R, Lane R, Lim P, et al. Efficacy and safety of fixed-dose esketamine nasal spray combined with a new oral antidepressant in treatment-resistant depression: results of a randomized, double-blind, active-controlled study (TRANSFORM-1). Int J Neuropsychopharmacol. 2019;22(10):616–30.
76. Ochs-Ross R, Daly EJ, Zhang Y, Lane R, Lim P, Morrison RL, et al. Efficacy and safety of esketamine nasal spray plus an oral antidepressant in elderly patients with treatment-resistant depression-TRANSFORM-3. Am J Geriatr Psychiatry. 2020;28(2):121–41.
77. Spravato Prescribing Information. Spravato (esketamine) [package insert]. Titusville, NJ: Janssen Pharmaceutical; 2019.
78. Anis NA, Berry SC, Burton NR, Lodge D. The dissociative anaesthetics, ketamine and phencyclidine, selectively reduce excitation of central mammalian neurones by N-methyl-aspartate. Br J Pharmacol. 1983;79(2):565–75.
79. Martin D, Lodge D. Ketamine acts as a non-competitive N-methyl-D-aspartate antagonist on frog spinal cord in vitro. Neuropharmacology. 1985;24(10):999–1003.
80. Yamamura T, Harada K, Okamura A, Kemmotsu O. Is the site of action of ketamine anesthesia the N-methyl-D-aspartate receptor? Anesthesiology. 1990;72(4):704–10.
81. Zanos P, Gould TD. Mechanisms of ketamine action as an antidepressant. Mol Psychiatry. 2018;23(4):801–11.
82. Andrade C. Ketamine for depression, 4: in what dose, at what rate, by what route, for how long, and at what frequency? J Clin Psychiatry. 2017;78(7):e852–7.
83. Duman RS, Li N, Liu RJ, Duric V, Aghajanian G. Signaling pathways underlying the rapid antidepressant actions of ketamine. Neuropharmacology. 2012;62(1):35–41.
84. Aleksandrova LR, Phillips AG, Wang YT. Antidepressant effects of ketamine and the roles of AMPA glutamate receptors and other mechanisms beyond NMDA receptor antagonism. J Psychiatry Neurosci. 2017;42(4):222–9.
85. Orser BA, Pennefather PS, MacDonald JF. Multiple mechanisms of ketamine blockade of N-methyl-D-aspartate receptors. Anesthesiology. 1997;86(4):903–17.
86. Yang Y, Cui Y, Sang K, Dong Y, Ni Z, Ma S, et al. Ketamine blocks bursting in the lateral habenula to rapidly relieve depression. Nature. 2018;554(7692):317–22.
87. Homayoun H, Moghaddam B. NMDA receptor hypofunction produces opposite effects on prefrontal cortex interneurons and pyramidal neurons. J Neurosci. 2007;27(43):11496–500.
88. Chowdhury GM, Zhang J, Thomas M, Banasr M, Ma X, Pittman B, et al. Transiently increased glutamate cycling in rat PFC is associated with rapid onset of antidepressant-like effects. Mol Psychiatry. 2017;22(1):120–6.

89. Miller OH, Yang L, Wang CC, Hargroder EA, Zhang Y, Delpire E, et al. GluN2B-containing NMDA receptors regulate depression-like behavior and are critical for the rapid antidepressant actions of ketamine. elife. 2014;3:3581.
90. Ignácio ZM, Réus GZ, Arent CO, Abelaira HM, Pitcher MR, Quevedo J. New perspectives on the involvement of mTOR in depression as well as in the action of antidepressant drugs. Br J Clin Pharmacol. 2016;82(5):1280–90.
91. Cavalleri L, Merlo Pich E, Millan MJ, Chiamulera C, Kunath T, Spano PF, et al. Ketamine enhances structural plasticity in mouse mesencephalic and human iPSC-derived dopaminergic neurons via AMPAR-driven BDNF and mTOR signaling. Mol Psychiatry. 2018;23(4): 812–23.
92. Li N, Lee B, Liu RJ, Banasr M, Dwyer JM, Iwata M, et al. mTOR-dependent synapse formation underlies the rapid antidepressant effects of NMDA antagonists. Science. 2010;329(5994):959–64.
93. Abdallah CG, Averill LA, Gueorguieva R, Goktas S, Purohit P, Ranganathan M, et al. Modulation of the antidepressant effects of ketamine by the mTORC1 inhibitor rapamycin. Neuropsychopharmacology. 2020;45(6):990–7.
94. Suzuki K, Monteggia LM. The role of eEF2 kinase in the rapid antidepressant actions of ketamine. Adv Pharmacol. 2020;89:79–99.
95. Zarate CA Jr, Singh JB, Quiroz JA, De Jesus G, Denicoff KK, Luckenbaugh DA, et al. A double-blind, placebo-controlled study of memantine in the treatment of major depression. Am J Psychiatry. 2006;163(1):153–5.
96. Yamaguchi JI, Toki H, Qu Y, Yang C, Koike H, Hashimoto K, et al. (2R,6R)-Hydroxynorketamine is not essential for the antidepressant actions of (R)-ketamine in mice. Neuropsychopharmacology. 2018;43(9):1900–7.
97. Yang C, Kobayashi S, Nakao K, Dong C, Han M, Qu Y, et al. AMPA receptor activation-independent antidepressant actions of ketamine metabolite (S)-Norketamine. Biol Psychiatry. 2018;84(8):591–600.
98. Morris PJ, Moaddel R, Zanos P, Moore CE, Gould TD, Zarate CA Jr, et al. Synthesis and N-methyl-d-aspartate (NMDA) receptor activity of ketamine metabolites. Org Lett. 2017;19 (17):4572–5.
99. Pfenninger EG, Durieux ME, Himmelseher S. Cognitive impairment after small-dose ketamine isomers in comparison to equianalgesic racemic ketamine in human volunteers. Anesthesiology. 2002;96(2):357–66.
100. Bahji A, Vazquez GH, Zarate CA Jr. Comparative efficacy of racemic ketamine and esketamine for depression: a systematic review and meta-analysis. J Affect Disord. 2021;278:542–55.
101. Fukumoto K, Toki H, Iijima M, Hashihayata T, Yamaguchi JI, Hashimoto K, et al. Antidepressant potential of (R)-ketamine in rodent models: comparison with (S)-ketamine. J Pharmacol Exp Ther. 2017;361(1):9–16.
102. Fu DJ, Ionescu DF, Li X, Lane R, Lim P, Sanacora G, et al. Esketamine nasal spray for rapid reduction of major depressive disorder symptoms in patients who have active suicidal ideation with intent: double-blind, randomized study (ASPIRE I). J Clin Psychiatry. 2020;81(3): 19m13191. https://doi.org/10.4088/JCP.19m13191.
103. Ionescu DF, Fu DJ, Qiu X, Lane R, Lim P, Kasper S, et al. Esketamine nasal spray for rapid reduction of depressive symptoms in patients with major depressive disorder who have active suicide ideation with intent: results of a phase 3, double-blind, randomized study (ASPIRE II). Int J Neuropsychopharmacol. 2021;24(1):22–31.
104. Miller AH, Maletic V, Raison CL. Inflammation and its discontents: the role of cytokines in the pathophysiology of major depression. Biol Psychiatry. 2009;65(9):732–41.
105. Miller AH, Raison CL. The role of inflammation in depression: from evolutionary imperative to modern treatment target. Nat Rev Immunol. 2016;16(1):22–34.
106. Ader R, Cohen N. Behaviorally conditioned immunosuppression. Psychosom Med. 1975;37 (4):333–40.

107. Plaut M. Lymphocyte hormone receptors. Annu Rev Immunol. 1987;5:621–69.
108. Cserr HF, Harling-Berg CJ, Knopf PM. Drainage of brain extracellular fluid into blood and deep cervical lymph and its immunological significance. Brain Pathol. 1992;2(4):269–76.
109. Zorrilla EP, Luborsky L, McKay JR, Rosenthal R, Houldin A, Tax A, et al. The relationship of depression and stressors to immunological assays: a meta-analytic review. Brain Behav Immun. 2001;15(3):199–226.
110. Bierhaus A, Wolf J, Andrassy M, Rohleder N, Humpert PM, Petrov D, et al. A mechanism converting psychosocial stress into mononuclear cell activation. Proc Natl Acad Sci. 2003;100 (4):1920–5.
111. Pace TW, Mletzko TC, Alagbe O, Musselman DL, Nemeroff CB, Miller AH, et al. Increased stress-induced inflammatory responses in male patients with major depression and increased early life stress. Am J Psychiatry. 2006;163(9):1630–3.
112. Raison CL, Capuron L, Miller AH. Cytokines sing the blues: inflammation and the pathogenesis of depression. Trends Immunol. 2006;27(1):24–31.
113. Brydon L, Harrison NA, Walker C, Steptoe A, Critchley HD. Peripheral inflammation is associated with altered substantia nigra activity and psychomotor slowing in humans. Biol Psychiatry. 2008;63(11):1022–9.
114. Dowlati Y, Herrmann N, Swardfager W, Liu H, Sham L, Reim EK, et al. A meta-analysis of cytokines in major depression. Biol Psychiatry. 2010;67(5):446–57.
115. Kopra E, Mondelli V, Pariante C, Nikkheslat N. Ketamine's effect on inflammation and kynurenine pathway in depression: a systematic review. J Psychopharmacol. 2021;35(8): 934–45.
116. Kim H, Chen L, Lim G, Sung B, Wang S, McCabe MF, et al. Brain indoleamine 2,3-dioxygenase contributes to the comorbidity of pain and depression. J Clin Invest. 2012;122(8):2940–54.
117. Dobos N, de Vries EF, Kema IP, Patas K, Prins M, Nijholt IM, et al. The role of indoleamine 2,3-dioxygenase in a mouse model of neuroinflammation-induced depression. J Alzheimers Dis. 2012;28(4):905–15.
118. Müller N, Schwarz MJ. The immune-mediated alteration of serotonin and glutamate: towards an integrated view of depression. Mol Psychiatry. 2007;12(11):988–1000.
119. Borland LM, Michael AC. Voltammetric study of the control of striatal dopamine release by glutamate. J Neurochem. 2004;91(1):220–9.
120. Myint AM, Schwarz MJ, Müller N. The role of the kynurenine metabolism in major depression. J Neural Transm. 2012;119(2):245–51.
121. Stone TW, Perkins MN. Quinolinic acid: a potent endogenous excitant at amino acid receptors in CNS. Eur J Pharmacol. 1981;72(4):411–2.
122. Gibney SM, McGuinness B, Prendergast C, Harkin A, Connor TJ. Poly I:C-induced activation of the immune response is accompanied by depression and anxiety-like behaviours, kynurenine pathway activation and reduced BDNF expression. Brain Behav Immun. 2013;28:170–81.
123. Frommberger UH, Bauer J, Haselbauer P, Fräulin A, Riemann D, Berger M. Interleukin-6-(IL-6) plasma levels in depression and schizophrenia: comparison between the acute state and after remission. Eur Arch Psychiatry Clin Neurosci. 1997;247(4):228–33.
124. Yirmiya R, Pollak Y, Morag M, Reichenberg A, Barak O, Avitsur R, et al. Illness, cytokines, and depression. Ann N Y Acad Sci. 2000;917:478–87.
125. Rethorst CD, Toups MS, Greer TL, Nakonezny PA, Carmody TJ, Grannemann BD, et al. Pro-inflammatory cytokines as predictors of antidepressant effects of exercise in major depressive disorder. Mol Psychiatry. 2013;18(10):1119–24.
126. Jia Y, Liu L, Sheng C, Cheng Z, Cui L, Li M, et al. Increased serum levels of cortisol and inflammatory cytokines in people with depression. J Nerv Ment Dis. 2019;207(4):271–6.
127. Müller N, Schwarz MJ, Dehning S, Douhe A, Cerovecki A, Goldstein-Müller B, et al. The cyclooxygenase-2 inhibitor celecoxib has therapeutic effects in major depression: results of a

double-blind, randomized, placebo controlled, add-on pilot study to reboxetine. Mol Psychiatry. 2006;11(7):680–4.
128. Akhondzadeh S, Jafari S, Raisi F, Nasehi AA, Ghoreishi A, Salehi B, et al. Clinical trial of adjunctive celecoxib treatment in patients with major depression: a double blind and placebo controlled trial. Depress Anxiety. 2009;26(7):607–11.
129. Krause D, Myint AM, Schuett C, Musil R, Dehning S, Cerovecki A, et al. High Kynurenine (a tryptophan metabolite) predicts remission in patients with major depression to add-on treatment with Celecoxib. Front Psych. 2017;8:16.
130. Halaris A, Cantos A, Johnson K, Hakimi M, Sinacore J. Modulation of the inflammatory response benefits treatment-resistant bipolar depression: a randomized clinical trial. J Affect Disord. 2020;261:145–52.
131. Malaguarnera M, Di Fazio I, Restuccia S, Pistone G, Ferlito L, Rampello L. Interferon alpha-induced depression in chronic hepatitis C patients: comparison between different types of interferon alpha. Neuropsychobiology. 1998;37(2):93–7.
132. Musselman DL, Lawson DH, Gumnick JF, Manatunga AK, Penna S, Goodkin RS, et al. Paroxetine for the prevention of depression induced by high-dose interferon alfa. N Engl J Med. 2001;344(13):961–6.
133. Raison CL, Borisov AS, Majer M, Drake DF, Pagnoni G, Woolwine BJ, et al. Activation of central nervous system inflammatory pathways by interferon-alpha: relationship to monoamines and depression. Biol Psychiatry. 2009;65(4):296–303.
134. Schiepers OJ, Wichers MC, Maes M. Cytokines and major depression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2005;29(2):201–17.
135. Owens MJ, Nemeroff CB. Physiology and pharmacology of corticotropin-releasing factor. Pharmacol Rev. 1991;43(4):425–73.
136. Pariante CM, Miller AH. Glucocorticoid receptors in major depression: relevance to pathophysiology and treatment. Biol Psychiatry. 2001;49(5):391–404.
137. Claes SJ. CRH, stress, and major depression: a psychobiological interplay. Vitam Horm. 2004;69:117–50.
138. Holsboer F, Ising M. Central CRH system in depression and anxiety—evidence from clinical studies with CRH1 receptor antagonists. Eur J Pharmacol. 2008;583(2–3):350–7.
139. Musazzi L, Racagni G, Popoli M. Stress, glucocorticoids and glutamate release: effects of antidepressant drugs. Neurochem Int. 2011;59(2):138–49.
140. van Tol MJ, van der Wee NJ, van den Heuvel OA, Nielen MM, Demenescu LR, Aleman A, et al. Regional brain volume in depression and anxiety disorders. Arch Gen Psychiatry. 2010;67(10):1002–11.
141. Loix S, De Kock M, Henin P. The anti-inflammatory effects of ketamine: state of the art. Acta Anaesthesiol Belg. 2011;62(1):47–58.
142. Van der Linden P, Gilbart E, Engelman E, Schmartz D, de Rood M, Vincent JL. Comparison of halothane, isoflurane, alfentanil, and ketamine in experimental septic shock. Anesth Analg. 1990;70(6):608–17.
143. Yli-Hankala A, Kirvelä M, Randell T, Lindgren L. Ketamine anaesthesia in a patient with septic shock. Acta Anaesthesiol Scand. 1992;36(5):483–5.
144. Lange M, Bröking K, van Aken H, Hucklenbruch C, Bone HG, Westphal M. Role of ketamine in sepsis and systemic inflammatory response syndrome. Anaesthesist. 2006;55(8):883–91.
145. Chen MH, Li CT, Lin WC, Hong CJ, Tu PC, Bai YM, et al. Rapid inflammation modulation and antidepressant efficacy of a low-dose ketamine infusion in treatment-resistant depression: a randomized, double-blind control study. Psychiatry Res. 2018;269:207–11.
146. Kiraly DD, Horn SR, Van Dam NT, Costi S, Schwartz J, Kim-Schulze S, et al. Altered peripheral immune profiles in treatment-resistant depression: response to ketamine and prediction of treatment outcome. Transl Psychiatry. 2017;7(3):1065.
147. Dale O, Somoyogi AA, Yiba L, Sullivan T, Shavit Y. Does intraoperative ketamine attenuate inflammatory reactivity following surgery? A systematic review and meta-analysis. Anesth Analg. 2012;115(4):934–43.

148. Haroon E, Daguanno AW, Woolwine BJ, Goldsmith DR, Baer WM, Wommack EC, et al. Antidepressant treatment resistance is associated with increased inflammatory markers in patients with major depressive disorder. Psychoneuroendocrinology. 2018;95:43–9.
149. Zhang GF, Wang J, Han JF, Guo J, Xie ZM, Pan W, et al. Acute single dose of ketamine relieves mechanical allodynia and consequent depression-like behaviors in a rat model. Neurosci Lett. 2016;631:7–12.
150. Verdonk F, Petit AC, Abdel-Ahad P, Vinckier F, Jouvion G, de Maricourt P, et al. Microglial production of quinolinic acid as a target and a biomarker of the antidepressant effect of ketamine. Brain Behav Immun. 2019;81:361–73.
151. Wang W, Liu L, Yang X, Gao H, Tang QK, Yin LY, et al. Ketamine improved depressive-like behaviors via hippocampal glucocorticoid receptor in chronic stress induced- susceptible mice. Behav Brain Res. 2019;364:75–84.
152. Johnston CJ, Fitzgerald PJ, Gewarges JS, Watson BO, Spencer-Segal JL. Ketamine decreases HPA axis reactivity to a novel stressor in male but not female mice. bioRxiv. 2021.
153. Walker AJ, Foley BM, Sutor SL, McGillivray JA, Frye MA, Tye SJ. Peripheral proinflammatory markers associated with ketamine response in a preclinical model of antidepressant-resistance. Behav Brain Res. 2015;293:198–202.
154. Rong C, Park C, Rosenblat JD, Subramaniapillai M, Zuckerman H, Fus D, et al. Predictors of response to ketamine in treatment resistant major depressive disorder and bipolar disorder. Int J Environ Res Public Health. 2018;15(4):771.
155. National Institute of Health. Clinicaltrials.gov: treatment resistant depression [Internet]. 2021. Clinicaltrials.gov.

# The Strategy of Targeting Peroxisome Proliferator-Activated Receptor (PPAR) in the Treatment of Neuropsychiatric Disorders

22

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**Abstract**

Peroxisome proliferator-activated receptors (PPARs) are nonsteroid nuclear receptors and transcription factors that regulate several neuroinflammatory and metabolic processes, recently involved in several neuropsychiatric conditions, including Alzheimer's disease, Parkinson's disease, major depressive disorder, post-traumatic stress disorder (PTSD), schizophrenia spectrum disorders, and autism spectrum disorders. PPARs are ligand-activated receptors that, following stimulation, induce neuroprotective effects by decreasing neuroinflammatory processes through inhibition of the nuclear factor kappa-light-chain-enhancer of activated B cell (NF-κB) expression and consequent suppression of pro-inflammatory cytokine production. PPARs heterodimerize with the retinoid X-receptor (RXR) and bind to PPAR-responsive regulatory elements (PPRE) in the promoter region of target genes involved in lipid metabolism, synthesis of cholesterol, catabolism of amino acids, and inflammation. Interestingly, PPARs are considered functionally part of the extended endocannabinoid (eCB) system that includes the classic eCB, anandamide, which act at cannabinoid receptor types 1 (CB1) and 2 (CB2) and are implicated in the pathophysiology of stress-related neuropsychiatric disorders. In preclinical studies, PPAR stimulation improves anxiety and depression-like behaviors by enhancing neurosteroid biosynthesis. The peculiar functional role of PPARs by exerting anti-inflammatory and neuroprotective effects and their expression localization in neurons and glial cells of corticolimbic circuits make them particularly interesting as novel

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therapeutic targets for several neuropsychiatric disorders characterized by underlying neuroinflammatory/neurodegenerative mechanisms. Herein, we discuss the pathological hallmarks of neuropsychiatric conditions associated with neuroinflammation, as well as the pivotal role of PPARs with a special emphasis on the subtype alpha (PPAR-α) as a suitable molecular target for therapeutic interventions.

**Keywords**

Alzheimer's disease · Schizophrenia · Depression · PTSD · Emotional behaviors · Neurosteroids · Endocannabinoid system · PPAR

## 22.1 Introduction

Chronic low-grade systemic inflammation and consequent activation of the pro-inflammatory response within the CNS, generally referred as *neuroinflammation*, represent a well-known pathogenetic mechanism affecting several neuropsychiatric conditions including Alzheimer's disease and Parkinson's disease, major depressive disorder (MDD), post-traumatic stress disorder (PTSD), autism spectrum disorder (ASD), and schizophrenia spectrum disorders (SSD) [1–6]. Abnormal activation of neuroinflammatory processes strongly associates with the severity of the disease progression and affects the treatment response [7, 8]. Peripheral inflammatory stimuli reach the brain and trigger astrocytes and microglia activation [9, 10]. Microglia cells constitute the innate immune cells and in the absence of inflammation are generally in a surveillant state [11, 12]. Moreover, triggered astrocytes can transform into a pro-inflammatory phenotype and regulate nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB) expression [13] and the production of pro-inflammatory cytokines through the toll-like receptor (TLR)4, which then stimulates microglia activation [14–16]. A microglia-neuron crosstalk via pro-inflammatory molecular mediators leads to neuroinflammation and neurodegeneration [2, 4, 17], which are both involved in the pathogenesis of neuropsychiatric conditions. Neuroinflammation causes abnormalities in synaptic plasticity with consequent abnormal neurotransmitter release, learning and memory process alterations, and ultimately neuronal death [18–20]. Under physiological conditions, microglia play an active role in defending the central nervous system from noxious stimuli via an active surveillance and promptly respond to challenging events that cause damage to neuronal cells. This occurs through activation of a cascade of inflammatory processes mediated by TLR signalling pathway that converges to the NF-κB transcription factor and pro-inflammatory cytokine production. Besides the underlying pathological conditions, associated risk factors including alcohol, drugs of abuse, stress, and infections, common among mood disorders and schizophrenia populations, activate neuroinflammatory processes. For example, abnormal expression of TLR has been reported in the early stages of schizophrenia and Alzheimer's disease and linked to cognitive deficits [21–23]. Systemic

inflammation can also affect the brain and its development as hypothesized for ASD vulnerability [24, 25].

The nuclear receptor peroxisome proliferator-activated receptor (PPAR) is involved in metabolic syndrome pathophysiology and several other inflammatory-based conditions, including neuropsychiatric disorders [26, 27]. For instance, several studies have recently revealed its role in the inflammatory mechanisms leading to Alzheimer's disease and MDD as well as the intriguingly pharmacological strategy of targeting PPAR in the treatment of several neuropsychiatric disorders.

Hereinafter, we will review the functional and therapeutic role of PPARs by exerting anti-inflammatory and neuroprotective effects underlying neurodevelopment and mood disorders. We will also discuss their expression localization across neurons and glial cells of corticolimbic areas involved in behavior regulation in several neuropsychiatric disorders. The pivotal role of PPAR-α as an emerging molecular target for the treatment of neuroinflammatory and neurodegenerative diseases will also be analyzed.

### 22.1.1 Peroxisome Proliferator-Activated Receptor (PPAR)-α and Inflammation

PPARs belong to the class of nonsteroid nuclear receptors with transcription factor activity, and consist of three isoforms, α, β/δ, and γ, encoded by NR1C1, NR1C2, and NR1C3 gene respectively, and differ for target genes, physiological functions, and tissue distribution [28, 29]. PPARs are expressed in many cellular types that exhibit differences in ligand specificity and activation of metabolic pathways.

PPAR-α and PPAR-δ are preferentially involved in the control of β-oxidation in organs with high energy demands like the heart, skeletal muscle, liver, and kidneys, whereas PPAR-γ is highly expressed in peripheral tissues with a high fatty acid synthesis and storage such as the adipose tissue [30]. All PPAR isotypes are expressed in the CNS in both neurons and glia with a unique pattern of brain areas and cell-type expression [31]. It has been reported that PPAR-α colocalizes with neurons, astrocytes, and microglia, PPAR-β/PPAR-δ colocalizes with neurons and astrocytes in white matter but not microglia, and PPAR-γ colocalizes with neurons and astrocytes but not microglia in the human brain. Also, PPAR-α is the only isotype to colocalize with all cell types in both adult mouse and adult human brain [32, 33], making this specific isotype a suitable target for pathological conditions that involve a glia-neuron crosstalk network. PPAR-α heterodimerizes with retinoid X receptors (RXR) to bind DNA-responsive elements (PPREs) on targeted gene promoters and, thereby, regulates transcription of multiple genes [31, 34]. Recently, we studied PPAR-α expression and its epigenetic regulation in a mouse model of stress-related disorders. We observed that *Ppar*-α gene promoter was hypermethylated in the hippocampus of socially isolated mice associated with a decrease of its mRNA expression and increased proinflammatory markers [35]. To our knowledge, this finding contributes the first demonstration that epigenetic changes occur at the *Ppar-α* gene promoter in the adult mouse brain.

The role of PPAR-α in brain proinflammatory processes and degeneration has become an emerging concept that has been intensively investigated in the past few years. PPAR-α exhibits anti-inflammatory effects and neuroprotective activity by modulating the expression of proinflammatory mediators, including the enzyme complex IkappaB kinase (IκB), an NF-κB inhibitor involved in propagating the cellular inflammatory response [36].

A number of studies have shown that the stimulation of PPAR-α mediated by the endogenous ligand palmitoylethanolamide (PEA) exerts anti-inflammatory effects through the inhibition of NF-kB signalling [36, 37]. PEA also potentiates neurosteroid biosynthesis and improves behavioral deficits induced by protracted stress in rodents [38, 39]. PEA is an endocannabinoid (eCB)-like bioactive lipid mediator, primarily targeting PPAR-α, with pleiotropic effects including anti-inflammatory, analgesic, anticonvulsant, antimicrobial, antipyretic, antiepileptic, immunomodulatory, and neuroprotective functions [40–42]. PEA's pleiotropic effects create potential therapeutic benefits in many pathological conditions, including neuroinflammatory and neurodegeneration [43, 44]. Systemic administration of PEA in socially isolated mice reversed the stress-induced downregulation of PPAR-α expression in the brain [39]. PEA also reversed the affective-like behavior by increasing neurosteroid biosynthesis and probably by enhancing the anti-inflammatory component associated with this mechanism. Indeed, chronic stress-induced hypermethylation of PPAR-α (Fig. 22.1) was associated with an increased pro-inflammatory response investigated in the hippocampus of socially

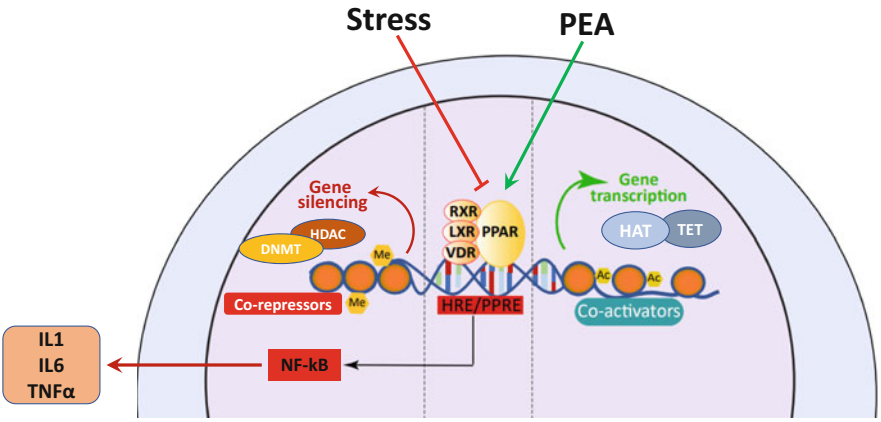


**Fig. 22.1** Schematic representation of stress effects on PPAR expression and regulation of target genes. Stress increases inflammation mediated through NF-kB pathway activation and TNF release. By releasing NF-kB-dependent inflammatory markers, including IL1, IL6, and TNF-α, astrocytes become reactive and crosstalk with activated microglia, ultimately leading to neuronal damage. PPAR-α is known to exhibit anti-inflammatory effects and a neuroprotective activity by modulating the expression of inflammatory mediators, including IκB, an NF-κB inhibitor, leading to the suppression of cytokine production. Stress induces epigenetic changes that alter PPAR-α expression, including hypermethylation of Ppar-α gene promoter [35], and these effects are associated with increased inflammation and altered behavior

isolated mice [35]. These preliminary findings strongly support our hypothesis of a beneficial role of PEA in the treatment of neuroinflammatory-based neuropsychiatric disorders.

PPAR-α expressed in astrocytes and neurons is deeply involved in several physiological and pathological conditions, including regulation of mitochondrial and proteasomal function, neuroinflammation, oxidative stress, and neurodegeneration, which are considered key pathogenetic mechanisms involved in stress-related disorders such as anxiety spectrum disorders and depression [27, 38, 39]. Deficits in PPAR-α signalling have also been observed in elevated neuroinflammatory processes in schizophrenia pathogenesis [45, 46]. Accordingly, a therapeutic role for PPAR-α ligands includes treatment of neurodegenerative disorders and addiction [47–49].

Pro-inflammatory markers, such as NFκB, PGE2, iNOS, and COX-2, are highly expressed in a cohort of schizophrenia patients [50, 51], compared to healthy controls. This supports the hypothesis that targeting these molecular players may represent a novel therapeutic strategy for schizophrenia management [52]. Moreover, it is well established that PPARs elicit anti-inflammatory effects via inhibition of the NF-κB pathway, and its popularity among the scientific community as a target for the treatment of neuroinflammatory conditions, including schizophrenia, is fast-growing. The finding that PPAR-α is target for endogenous ligands to induce neurosteroid biosynthesis [38] further supports the hypothesis that activation of PPAR-α may prevent inflammatory-induced neuronal damage by upregulating neurosteroid metabolites with anti-inflammatory actions [53–55].

Administration with PPAR agonists in the knockdown of GluN1, an NMDA receptor subunit, model of schizophrenia showed positive results in long-term memory improvement [56]. In the schizophrenia animal model of maternal immune activation (MIA), prenatal treatment with the PPAR-α agonist fenofibrate attenuates the MIA-induced biochemical and behavioral deficits [57]. Favorable effects were also observed in the management of the second-generation antipsychotic olanzapine-induced weight gain by using a histamine agonist with PPAR-α modulating activity [58]. Interestingly, besides endogenous and synthetic compounds, such as the fibrates, several natural bioactive compounds and phytocannabinoids act as ligands for PPAR-α including polyphenols and unsaturated fatty acids which stimulate its expression and induce anti-inflammatory effects [28, 59, 60], supporting a relevant role for natural bioactive compounds found in functional foods that can be added to the diet of these populations.

Also, PPAR-γ ligands have been studied for their role in cognition [49], which is a core symptom of schizophrenia. Taken together, these findings suggest that PPAR activation can be investigated in a broad spectrum of conditions associated with mood, neurodevelopmental, and neurodegenerative disorders that are characterized by pervasive neuroinflammation. Here, we discuss the anti-inflammatory role of PPAR-α in (1) neurodegenerative disorders such as Alzheimer's disease and Parkinson's disease; (2) psychiatric disorders such as schizophrenia spectrum disorders, major depressive disorder, and PTSD; and (3) neurodevelopmental disorders such as autism spectrum disorders (ASD).

## 22.2 PPAR-α as Potential Molecular Target for Neuropsychiatric Conditions

### 22.2.1 Alzheimer's Disease

Alzheimer's disease represents the most common cause of dementia; accounting for an estimated 60–70% of cases worldwide [61]. Although the pathological hallmarks of Alzheimer's disease in the brain are amyloid-β (Aβ) plaques and abnormal tau tangles, which were extensively addressed by research in the last decades, these abnormalities did not explain the persistence of cognitive decline despite the patients' amyloid load was reduced in clinical trials [62, 63]. The recent evidence of increased levels of pro-inflammatory markers in subjects with Alzheimer's disease and the identification of Alzheimer's disease risk genes associated with innate immune functions strongly supported the hypothesis of neuroinflammation as a prominent mechanism involved in the pathogenesis of Alzheimer's disease [64, 65]. Neuroinflammation consists in an inflammatory response within the CNS usually caused by various pathological stimuli such as infections, traumatic brain injuries, stroke, and toxins [66] as well as chronic stress [67]. The neuroinflammatory mediators are produced and released by activated innate immune cells represented by the resident CNS glia cells (microglia and astrocytes) and endothelial cells or derived from peripheral immune cells. Importantly, the inflammatory response may lead to synaptic dysfunction, neuronal death, and inhibition of neurogenesis contributing to the neurodegeneration and cognitive deterioration in Alzheimer's disease [68]. Our lab just recently demonstrated that protracted social isolation stress induced an increase in pro-inflammatory markers in the hippocampus associated with epigenetic changes in PPAR-α gene promoter and consequent suppression of its expression [35]. Decreased PPAR-α expression has also been implicated in the pathogenesis of Alzheimer's disease [69], and administration of PPAR-α agonists (gemfibrozil and WY14643) decreases amyloid pathology and reverses memory deficits and anxiety symptoms in APP-PSEN1ΔE9 mice [70] suggesting that PPAR-α activation may be relevant for the improvement of cognitive function [71].

### 22.2.2 Parkinson's Disease

Parkinson's disease is the most invalidating motor disorder characterized by rest tremor, rigidity, and bradykinesia [72]. Motor symptoms usually represent the "core" feature of the disease although non-motor symptoms are also frequent and include autonomic dysfunction and cognitive and psychiatric changes such as major depressive disorder and psychosis [73–75]. Parkinson's disease is a neurodegenerative disorder caused by loss of dopaminergic neurons projecting from the substantia nigra pars compacta to the caudate nucleus and putamen (striatum) associated with mitochondrial dysfunction, oxidative stress, excitotoxicity, apoptosis, and inflammation [76–78]. Animal models for Parkinson's disease have been reproduced by

exposure to the toxin 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP), which induces degeneration of dopaminergic neurons in the substantia nigra [79]. In MPTP-exposed rodents, the PPAR-γ activator pioglitazone protects against neurotoxicity, decreasing microglial activation, and iNOS-positive cells, as well as inhibiting monoamine oxidase-B expression [80]. In a chronic MPTP model, administration of the PPAR-γ agonist rosiglitazone protected from dopaminergic neuron loss [81]. Similar effects were obtained with MHY908, a PPAR-α/PPAR-γ dual agonist, that showed neuroprotective effects by reducing microglial activation and neuroinflammation in the MPTP mouse model [82]. In addition, studies showed that the PPAR-α agonist fenofibrate decreases the toxicity induced by MPTP [83] by reducing the production of inflammatory cytokines [84]. Taken together, these findings strongly suggest that targeting PPARs may change the Parkinson's disease trajectory and could be a pertinent target for Parkinson's disease treatment.

### 22.2.3 Schizophrenia Spectrum Disorders

Schizophrenia spectrum disorder is a severe chronic, highly heterogenic, neurodevelopmental disorder associated with progressive neuronal loss, brain structural and functional changes, and a significant shortened life expectancy [85–87]. Although several hypotheses have been made including the dopaminergic hypothesis and the GABAergic/glutamatergic neurotransmission imbalance hypothesis, the causes of schizophrenia are not completely elucidated. In fact, besides the individual genetic vulnerability, epigenetic changes occurring as consequence of environmental adverse events ("second hit"), including stress, drug abuse, and trauma, play an important role in alterations in cortical GABAergic/glutamatergic neurotransmission [88, 89]. Recently, neuroinflammatory processes in schizophrenia have been investigated and demonstrated by the evidence that individuals affected by schizophrenia show elevated expression of inflammatory markers such as interleukin (IL)-β, IL-6, or C-reactive protein (CRP) in both the brain and peripheral blood [90–92]. Recently, it has been shown that peripheral low-grade inflammation is associated with ultra-resistance to treatment in schizophrenia UTRS [93–95], suggesting that inflammation contributes to treatment resistance. A study reported that schizophrenia patients show a specific immune-inflammatory biomarker pattern characterized by increased NFκB, PGE2, iNOS, and COX-2 levels compared to bipolar disorder patients and healthy controls [6, 96], hypothesizing that pharmacological modulation of these inflammatory markers may constitute a promising therapeutic target. Indeed, PPAR stimulation plays a pivotal role in schizophrenia illness severity and treatment response by modulating pro-inflammatory cytokine expression.

The mechanism that links schizophrenia to inflammation relies on the evidence that systemic inflammation caused by different pathological conditions leads to pro-inflammatory cytokine release into the general system, which then creates a mirror inflammatory response in the brain via microglia activation and secondary production of pro-inflammatory cytokines, such as tumor necrosis factor (TNF), IL-1β, and

IL-6 [52, 93, 97, 98]. Neuroinflammation during development or early life, in turn, alters neuronal maturation and synaptic plasticity by altering the glutamatergic system [45, 99, 100]. Through microglia-neuron crosstalk, pro-inflammatory cytokines, including TNF-α, which alters the cell membrane expression of AMPA and NMDA glutamate receptors leading to abnormal calcium mobilization, stimulates glutamate release that precipitates excitotoxicity processes, inhibits glutamate transport on astrocytes, and alters $GABA_A$ receptor expression [101]. PPAR-α exhibits anti-inflammatory effects and neuroprotective activity by modulating the expression of inflammatory mediators, such as IκB, an NF-κB inhibitor [36]. Thus, PPAR-α activation may reduce (1) illness severity and progression, (2) metabolic adverse effects induced by certain medications (i.e., antipsychotics), and (3) treatment response. To this end, PPAR-α stimulation may represent a useful anti-inflammatory tool for therapeutic intervention in schizophrenia in which neuroinflammation plays a pivotal role.

An additional mechanism that has emerged recently highlights the role of neurosteroids, including allopregnanolone, in neuronal development and brain functioning by modulating GABAergic neurotransmission via a direct action on mainly extra-synaptic $GABA_A$ receptors [102, 103]. Also, sulfated neurosteroids modulate *N*-methyl-d-aspartate (NMDA) receptors, a key molecular player in schizophrenia pathogenesis, with different electrophysiological effects and binding affinity based on the NMDA receptor subunit composition. For instance, pregnanolone sulfate inhibits tonic-mediated NMDA neurotransmission, which provides neuroprotection [104, 105]. PPAR-α stimulation by systemic administration of PEA promotes allopregnanolone and its sulfated congener biosynthesis and therefore might improve schizophrenia course by reducing the impact of inflammation on neuronal damage.

Treatment-resistant schizophrenia (TRS) represents a subgroup of population that does not respond to at least two first-line antipsychotic medications at adequate dose and duration with documented compliance [106]. TRS affects around 30% of subjects with schizophrenia and results in a high risk of relapses, poor global functioning, social impairments including unemployment, and reduced overall quality of life [107]. The only approved evidence-based treatment for TRS is the atypical antipsychotic clozapine, which is still considered the most effective antipsychotic but, unfortunately, used only as a second-line treatment due to its adverse effects and tolerability profile [108]. The biological mechanisms underlying the neuropathology of TRS are not completely understood. Recent evidence highlights the role of several conditions including dopamine supersensitivity, hyperdopaminergic and normodopaminergic subtypes, glutamate dysregulation, inflammation and oxidative stress, and serotonin dysregulation [109]. Recently, Keller et al. (2018) reported that treatment strategies using anti-inflammatory agents showed some benefits in people with schizophrenia [110].

Also, only 40% of treatment-resistant schizophrenia subpopulation responds to clozapine [111], thus creating a subgroup of population (12–20%) that is clozapine-resistant and therefore referred to as *ultra-resistant* (UTRS) or clozapine-resistant schizophrenia (CRS) [112]. Although the pathophysiological bases of ultra-resistant

schizophrenia remain largely unknown, recent evidence suggests that UTRS is associated with chronic peripheral low-grade inflammation as resulted by increased levels of high-sensitivity C-reactive protein (hs-CRP) [93, 113]. Also, peripheral inflammatory markers correlate with poor treatment response prediction to antipsychotics [114, 115]. Given the remarkable pro-inflammatory component of this disorder, while it has not been experimented yet, it would be of interest to test whether PPAR agonists would be beneficial in this treatment-resistant population.

### 22.2.4 Autism Spectrum Disorder (ASD)

ASD is a neurodevelopmental disorder of childhood characterized by impaired social communication and social interaction and restricted and repetitive behaviors with a complex, multifactorial etiology [116]. The pathogenesis of ASD is characterized by a high heterogeneity and associated with a strong genetic vulnerability combined with environmental adverse factors in the early phase of development [117, 118]. Current FDA-approved medications for ASD belong to the class of antipsychotics (e.g., risperidone and aripiprazole) that mainly target irritability and aggressive behaviors in children with ASD, but these drugs fail to treat core symptoms of ASD. Although alterations in GABAergic/glutamatergic neurotransmission associated with abnormal synaptic plasticity are strongly involved in the pathogenesis of ASD, the precise mechanisms remain unclear. The growing evidence suggests a relevant role for systemic inflammatory dysregulation and neuroinflammation in the neuropathology of ASD [119–122]. PPARs regulate inflammatory pathways by controlling gene expression of key transcription factors such as NF-κB or cyclooxygenase. Activation of PPAR-α and PPAR-γ by selective agonists has been reported to exert neuroprotective effects by reducing oxidative stress and neuroinflammation, which are processes involved in ASD pathophysiology [123].

Clinical studies showed a significant reduction in irritability, lethargy, stereotypy, and hyperactivity with the PPAR-γ agonist pioglitazone [124]. In a 10-week study, pioglitazone was also studied as add-on strategy to risperidone in the treatment of irritability in ASD [125]. Pioglitazone also favorably modifies behavioral symptoms along with a significant reduction of pro-inflammatory IL-6 [126].

Consistent with PPAR-α activation, neurobehavioral and biochemical benefits in an ASD animal model were observed following administration with fenofibrate that resulted in reduced oxidative stress and inflammation in several brain regions [127].

PPAR-α is required for normal cerebral functions, and its genetic ablation leads to repetitive behaviors and cognitive inflexibility in mice [128]. In another rodent model of ASD, the BTBR T + tf/J (BTBR) mouse, PEA reverted the altered phenotype and improved ASD-like behavior through PPAR-α activation. This effect was accompanied by decreased levels of inflammatory cytokines and restored the hippocampal brain-derived neurotrophic factor (BDNF) signaling pathway in BTBR mice [129]. Indeed, PPAR-α activation modulates BDNF expression and neuroplasticity [130], an action that was associated with improvement of mood

deficits [131]. Treatment with the selective PPAR-β/δ agonist GW0742 improved repetitive behaviors and lowered thermal sensitivity responses in the BTBR rodents while decreasing pro-inflammatory and increasing anti-inflammatory cytokines [132].

### 22.2.5 Major Depressive Disorder

Neuroinflammation has been implicated in the pathophysiology of depression [133, 134]. Dysregulations of immune system occurs in depressed patients and obstruct favorable prognosis and antidepressant treatment responses [135]. Synthetic agonists of PPAR-α have been investigated in clinical trials for their ability to improve depression symptoms [26]. PPARs are key molecular regulators of cell metabolism, energy homeostasis, cellular development, and differentiation; thus, their ligands find several clinical applications such as hyperlipidemia and hypertriglyceridemia in combination with statins, type 2 diabetes mellitus, metabolic syndrome, and nonalcoholic fatty liver disease, as well as neurodegenerative diseases, cancers, and inflammatory diseases where they play a relevant role between metabolic disorders and chronic low-grade systemic inflammation promoting anti-inflammatory effects [136]. In addition, PPAR agonists have been tested in mood disorders, especially major depression and depressive episodes in bipolar disorder, showing a marked improvement of depressive symptoms [137, 138], although the underlying mechanisms are not fully understood. We propose that PPARs might work in synergism with neurosteroids to exert their beneficial effects to relieve depressive symptoms. Interestingly, recent preclinical evidence suggest that PPAR-α activation induced by the administration of its endogenous modulator, PEA, induces biosynthesis of allopregnanolone in corticolimbic brain areas including the frontal cortex, hippocampus, and amygdala [38]. The PEA-induced neurosteroid increase was also correlated with behavioral dysfunction improvement expressed in a mouse model for PTSD and depression. Previous work showed that the PEA-induced activation of peripherally expressed PPAR-α also results in allopregnanolone upregulation [139, 140]. Altogether, these findings highlight the potential role of PPAR-α regulation as a suitable target for developing new strategies for the treatment of mood disorders that are characterized by deficiency in neurosteroidogenesis and elevated neuroinflammatory processes, including major depressive disorder, PTSD, and postpartum depression. Importantly, synthetic PPAR-α agonists, such as the fibrates, fenofibrate and clofibrate, which are FDA-approved for the treatment of hypercholesterolemia, could be repurposed to treat mood disorders by targeting the PPAR-allopregnanolone pathway [141, 142]. By this mechanism, enhancing allopregnanolone biosynthesis would enhance extrasynaptic $GABA_A$ receptor-mediated inhibitory signaling as well as decrease the pro-inflammatory pathway mediated by TLR4 activation [54, 55].

It has been recently reported that approximately one third of patients with major depression fail to reach even a partial response with current pharmacological treatments, and remission is obtained in only about one third of responders. Failure

to achieve a complete remission after trials with different antidepressants is known as treatment-resistant depression (TRD) [143–145]. Inflammation has been considered as potential underlying pathological mechanism in TRD [146, 147]. Increased levels of cytokines in depression provide the rationale for targeting the immune inflammatory system in MDD [148]. Treatment-resistant depression has been associated with increased peripheral levels of CRP in humans [149]. Moreover, inflammation is associated with depression, especially in women, and levels of CRP and interleukin (IL)-6 correlate with the response to antidepressants [150]. The anti-inflammatory celecoxib and some nonsteroidal anti-inflammatory agents (NSAIDs) improve depressive symptoms in adult subjects with major depressive disorder in combination with antidepressants [151, 152]. Similar results were obtained with the use of the TNF antagonist infliximab [153]. The process of neuroinflammation can also lead to neurodegeneration and alterations of neurosteroid synthesis [154]. Endogenous allopregnanolone in the brain is downregulated in rodent stress models and in humans with major depression. Previous studies showed that brain allopregnanolone levels fell markedly in rodents following protracted social isolation stress as well and in the bulbectomized model of depression [104, 155–157]. Interestingly, in vivo treatment with PEA ameliorated the behavioral phenotype in socially isolated mice [38, 39, 104] unveiling a potential novel mechanism as target for drug therapy in treatment-resistant depression.

### 22.2.6 Post-Traumatic Stress Disorder (PTSD)

PTSD is a debilitating trauma-related mental disorder that develops in individuals who experience or witness a life-threatening traumatic event [158]. Lifetime prevalence of PTSD is estimated at around 3.9% worldwide [3]. Diagnostic symptoms of PTSD include severe emotional distress or physical reactions, flashbacks, avoidance behavior, fear and anxiety, cognition impairment, mood changes, hyperarousal, and reactivity (irritability). Current available medications for symptom reduction include selective serotonin reuptake inhibitors (SSRIs) and serotonin-norepinephrine (SNRIs) such as fluoxetine, paroxetine, sertraline, and venlafaxine. Unfortunately, these current pharmacological treatments fail to address the immune-inflammatory responses and cognitive impairments as well as to achieve a complete remission, and augmentation strategies are needed [159]. Endocannabinoids, through the activation of CB1 and CB2 receptors, play an important role in the regulation of the amygdala-hippocampal-corticostriatal neuronal circuit involved in PTSD, specifically in stress response [160]. PEA recently has been proposed to be part of the extended eCB system inducing its main pharmacological effects by stimulating PPAR-α which plays a role in suppressing neuroinflammation and oxidative stress upon stimulation [38, 39], thereby linking the eCB system to inflammatory processes in the neurocircuit responsible for the PTSD pathogenesis. PPAR-α is decreased in the socially isolated mouse model for PTSD, and the treatment with PEA reverses this deficit causing an increase in PPAR-α expression and ameliorating the behavioral

phenotype, including improvement of fear extinction deficits and fear extinction retention [35, 38, 39].

Evidence showed a strong correlation of PTSD and alterations in oxidative stress, inflammation, and neurosteroid levels [161–166]. Individuals with PTSD exhibit significantly elevated blood levels of inflammatory markers, such as IL-1β, IL-6, TNF-α, and CRP, relative to healthy control subjects [167]. Mounting evidence indicates that the cannabinoid system is involved in multiple aspects of the pathophysiology of PTSD including inflammatory processes [168]. Although the potential risks of cannabis use for PTSD treatment outweigh the benefits [169], its anti-inflammatory properties might be beneficial in the treatment of PTSD and might open to new potential molecular targets involving the eCB system and extended eCB family by targeting PPAR-α. In fact, in the mouse model for PTSD induced by protracted social isolation, PPAR-α stimulation induced by PEA reverses the abnormal biochemical and behavioral phenotype [38, 39, 104], suggesting a therapeutic effect of PEA in PTSD symptoms potentially via stimulation of GABAergic neurotransmission and inflammatory processes inhibition [35].

### 22.2.7 Conclusions and Future Perspectives

PPARs represent novel fascinating targets for the treatment of neuropsychiatric disorders associated with elevated neuroinflammatory processes and neurodegeneration. Its epigenetic regulation in the brain under chronic stress conditions unveils new scenarios for drug therapy. A growing body of preclinical and clinical data suggests that neuroinflammation is a pathological process common to several neuropsychiatric conditions that deserves to be therapeutically addressed beyond the specific cause which characterizes the single disease. Based on the compelling scientific evidence provided, PPAR activation by downregulating neuroinflammation may help in the better management and treatment of neuropsychiatric disorders. Both natural or synthetic PPAR ligands, by stimulating anti-inflammatory mechanisms and, possibly, by enhancing neurosteroid biosynthesis, may provide a future treatment approach to prevent and alleviate pathophysiological processes that lead to the development of brain pathological conditions (Fig. 22.1).

**Potential Conflicts of Interests** GP is a paid consultant to PureTech Health, GABA Therapeutics, and NeuroTrauma Science and has two pending patent applications, one on N-palmitoylethanolamine (PEA) and peroxisome proliferator-activated receptor alpha (PPAR-α) agonists and one on allopregnanolone analogs in the treatment of neuropsychiatric disorders.

## References

1. Beurel E, Toups M, Nemeroff CB. The bidirectional relationship of depression and inflammation: double trouble. Neuron. 2020;107(2):234–56. https://doi.org/10.1016/j.neuron.2020.06.002.

2. Buckley PF. Neuroinflammation and schizophrenia. Curr Psychiatry Rep. 2019;21(8):72. https://doi.org/10.1007/s11920-019-1050-z.
3. Hori H, Kim Y. Inflammation and post-traumatic stress disorder. Psychiatry Clin Neurosci. 2019;73(4):143–53. https://doi.org/10.1111/pcn.12820.
4. Feigenson KA, Kusnecov AW, Silverstein SM. Inflammation and the two-hit hypothesis of schizophrenia. Neurosci Biobehav Rev. 2014;38:72–93. https://doi.org/10.1016/j.neubiorev.2013.11.006.
5. Inta D, Lang UE, Borgwardt S, Meyer-Lindenberg A, Gass P. Microglia activation and schizophrenia: lessons from the effects of minocycline on postnatal neurogenesis, neuronal survival and synaptic pruning. Schizophr Bull. 2017;43(3):493–6. https://doi.org/10.1093/schbul/sbw088.
6. Upthegrove R, Khandaker GM. Cytokines, oxidative stress and cellular markers of inflammation in schizophrenia. Curr Top Behav Neurosci. 2020;44:49–66. https://doi.org/10.1007/7854_2018_88.
7. Gonzalez-Burgos G, Lewis DA. NMDA receptor hypofunction, parvalbumin-positive neurons, and cortical gamma oscillations in schizophrenia. Schizophr Bull. 2012;38(5):950–7. https://doi.org/10.1093/schbul/sbs010.
8. Anderson G, Maes M. Schizophrenia: linking prenatal infection to cytokines, the tryptophan catabolite (TRYCAT) pathway, NMDA receptor hypofunction, neurodevelopment and neuroprogression. Prog Neuro-Psychopharmacol Biol Psychiatry. 2013;42:5–19. https://doi.org/10.1016/j.pnpbp.2012.06.014.
9. Notter T, Coughlin JM, Sawa A, Meyer U. Reconceptualization of translocator protein as a biomarker of neuroinflammation in psychiatry. Mol Psychiatry. 2018;23(1):36–47. https://doi.org/10.1038/mp.2017.232.
10. Kwon HS, Koh SH. Neuroinflammation in neurodegenerative disorders: the roles of microglia and astrocytes. Transl Neurodegener. 2020;9(1):42. https://doi.org/10.1186/s40035-020-00221-2.
11. Wolf SA, Boddeke HW, Kettenmann H. Microglia in physiology and disease. Annu Rev Physiol. 2017;79:619–43. https://doi.org/10.1146/annurev-physiol-022516-034406.
12. Colonna M, Butovsky O. Microglia function in the central nervous system during health and neurodegeneration. Annu Rev Immunol. 2017;35:441–68. https://doi.org/10.1146/annurev-immunol-051116-052358.
13. Linnerbauer M, Rothhammer V. Protective functions of reactive astrocytes following central nervous system insult. Front Immunol. 2020;11:573256. https://doi.org/10.3389/fimmu.2020.573256.
14. Sofroniew MV. Astrocyte barriers to neurotoxic inflammation. Nat Rev Neurosci. 2015;16(5):249–63. https://doi.org/10.1038/nrn3898.
15. Rosciszewski G, Cadena V, Murta V, Lukin J, Villarreal A, Roger T, Ramos AJ. Toll-like receptor 4 (TLR4) and triggering receptor expressed on myeloid Cells-2 (TREM-2) activation balance astrocyte polarization into a Proinflammatory phenotype. Mol Neurobiol. 2018;55(5):3875–88. https://doi.org/10.1007/s12035-017-0618-z.
16. Jha MK, Jo M, Kim JH, Suk K. Microglia-astrocyte crosstalk: an intimate molecular conversation. Neuroscientist. 2019;25(3):227–40. https://doi.org/10.1177/1073858418783959.
17. Szepesi Z, Manouchehrian O, Bachiller S, Deierborg T. Bidirectional microglia-neuron communication in health and disease. Front Cell Neurosci. 2018;12:323. https://doi.org/10.3389/fncel.2018.00323.
18. Rizzo FR, Musella A, De Vito F, Fresegna D, Bullitta S, Vanni V, Guadalupi L, Stampanoni Bassi M, Buttari F, Mandolesi G, Centonze D, Gentile A. Tumor necrosis factor and interleukin-1$\beta$ modulate synaptic plasticity during Neuroinflammation. Neural Plast. 2018;2018:8430123. https://doi.org/10.1155/2018/8430123.
19. Marinelli S, Basilico B, Marrone MC, Ragozzino D. Microglia-neuron crosstalk: signaling mechanism and control of synaptic transmission. Semin Cell Dev Biol. 2019;94:138–51. https://doi.org/10.1016/j.semcdb.2019.05.017.

20. Zheng ZH, Tu JL, Li XH, Hua Q, Liu WZ, Liu Y, Pan BX, Hu P, Zhang WH. Neuroinflammation induces anxiety- and depressive-like behavior by modulating neuronal plasticity in the basolateral amygdala. Brain Behav Immun. 2021;91:505–18. https://doi.org/10.1016/j.bbi.2020.11.007.
21. Crews FT, Lawrimore CJ, Walter TJ, Coleman LG Jr. The role of neuroimmune signaling in alcoholism. Neuropharmacology. 2017;122:56–73. https://doi.org/10.1016/j.neuropharm.2017.01.031.
22. Kéri S, Szabó C, Kelemen O. Antipsychotics influence toll-like receptor (TLR) expression and its relationship with cognitive functions in schizophrenia. Brain Behav Immun. 2017;62:256–64. https://doi.org/10.1016/j.bbi.2016.12.011.
23. Yang J, Wise L, Fukuchi KI. TLR4 cross-talk with NLRP3 Inflammasome and complement signaling pathways in Alzheimer's disease. Front Immunol. 2020;11:724. https://doi.org/10.3389/fimmu.2020.00724.
24. Jiang NM, Cowan M, Moonah SN, Petri WA Jr. The impact of systemic inflammation on neurodevelopment. Trends Mol Med. 2018;24(9):794–804. https://doi.org/10.1016/j.molmed.2018.06.008.
25. van Sadelhoff J, Perez Pardo P, Wu J, Garssen J, van Bergenhenegouwen J, Hogenkamp A, Hartog A, Kraneveld AD. The gut-immune-brain axis in autism spectrum disorders; a focus on amino acids. Front Endocrinol. 2019;10:247. https://doi.org/10.3389/fendo.2019.00247.
26. Cheng HS, Tan WR, Low ZS, Marvalim C, Lee J, Tan NS. Exploration and development of PPAR modulators in health and disease: an update of clinical evidence. Int J Mol Sci. 2019;20(20):5055. https://doi.org/10.3390/ijms20205055.
27. Tufano M, Pinna G. Is there a future for PPARs in the treatment of neuropsychiatric disorders? Molecules. 2020;25(5):1062. https://doi.org/10.3390/molecules25051062.
28. Grygiel-Górniak B. Peroxisome proliferator-activated receptors and their ligands: nutritional and clinical implications—a review. Nutr J. 2014;13:17. https://doi.org/10.1186/1475-2891-13-17.
29. Corrales P, Vidal-Puig A, Medina-Gómez G. PPARs and metabolic disorders associated with challenged adipose tissue plasticity. Int J Mol Sci. 2018;19(7):2124. https://doi.org/10.3390/ijms19072124.
30. Iglesias J, Morales L, Barreto GE. Metabolic and inflammatory adaptation of reactive astrocytes: role of PPARs. Mol Neurobiol. 2017;54(4):2518–38. https://doi.org/10.1007/s12035-016-9833-2.
31. Moreno S, Farioli-Vecchioli S, Cerù MP. Immunolocalization of peroxisome proliferator-activated receptors and retinoid X receptors in the adult rat CNS. Neuroscience. 2004;123(1):131–45. https://doi.org/10.1016/j.neuroscience.2003.08.064.
32. Heneka MT, Landreth GE. PPARs in the brain. Biochim Biophys Acta. 2007;1771(8):1031–45. https://doi.org/10.1016/j.bbalip.2007.04.016.
33. Warden A, Truitt J, Merriman M, Ponomareva O, Jameson K, Ferguson LB, Mayfield RD, Harris RA. Localization of PPAR isotypes in the adult mouse and human brain. Sci Rep. 2016;6:27618. https://doi.org/10.1038/srep27618.
34. Berger J, Moller DE. The mechanisms of action of PPARs. Annu Rev Med. 2002;53:409–35. https://doi.org/10.1146/annurev.med.53.082901.104018.
35. Matrisciano F, Pinna G. PPAR-α Hypermethylation in the hippocampus of mice exposed to social isolation stress is associated with enhanced neuroinflammation and aggressive behavior. Int J Mol Sci. 2021;22(19):10678. https://doi.org/10.3390/ijms221910678.
36. Bougarne N, Weyers B, Desmet SJ, Deckers J, Ray DW, Staels B, De Bosscher K. Molecular actions of PPARα in lipid metabolism and inflammation. Endocr Rev. 2018;39(5):760–802. https://doi.org/10.1210/er.2018-00064.
37. Chinetti G, Fruchart JC, Staels B. Peroxisome proliferator-activated receptors: new targets for the pharmacological modulation of macrophage gene expression and function. Curr Opin Lipidol. 2003;14(5):459–68. https://doi.org/10.1097/00041433-200310000-00006.

38. Locci A, Pinna G. Stimulation of peroxisome proliferator-activated receptor-α by N-Palmitoylethanolamine engages allopregnanolone biosynthesis to modulate emotional behavior. Biol Psychiatry. 2019;85(12):1036–45. https://doi.org/10.1016/j.biopsych.2019.02.006.
39. Locci A, Pinna G. Social isolation as a promising animal model of PTSD comorbid suicide: neurosteroids and cannabinoids as possible treatment options. Prog Neuro-Psychopharmacol Biol Psychiatry. 2019;92:243–59. https://doi.org/10.1016/j.pnpbp.2018.12.014.
40. D'Agostino G, Russo R, Avagliano C, Cristiano C, Meli R, Calignano A. Palmitoylethanolamide protects against the amyloid-β25-35-induced learning and memory impairment in mice, an experimental model of Alzheimer'sdisease. Neuropsychopharmacology. 2012;37(7):1784–92. https://doi.org/10.1038/npp.2012.25.
41. Scuderi C, Stecca C, Valenza M, Ratano P, Bronzuoli MR, Bartoli S, Steardo L, Pompili E, Fumagalli L, Campolongo P, Steardo L. Palmitoylethanolamide controls reactive gliosis and exerts neuroprotective functions in a rat model of Alzheimer's disease. Cell Death Dis. 2014;5(9):e1419. https://doi.org/10.1038/cddis.2014.376.
42. Clayton P, Hill M, Bogoda N, Subah S, Venkatesh R. Palmitoylethanolamide: a natural compound for health management. Int J Mol Sci. 2021;22(10):5305. https://doi.org/10.3390/ijms22105305.
43. Petrosino S, Di Marzo V. The pharmacology of palmitoylethanolamide and first data on the therapeutic efficacy of some of its new formulations. Br J Pharmacol. 2017;174(11):1349–65. https://doi.org/10.1111/bph.13580.
44. Beggiato S, Tomasini MC, Ferraro L. Palmitoylethanolamide (PEA) as a potential therapeutic agent in Alzheimer's disease. Front Pharmacol. 2019;10:821. https://doi.org/10.3389/fphar.2019.00821.
45. Na KS, Jung HY, Kim YK. The role of pro-inflammatory cytokines in the neuroinflammation and neurogenesis of schizophrenia. Prog Neuro-Psychopharmacol Biol Psychiatry. 2014;48:277–86. https://doi.org/10.1016/j.pnpbp.2012.10.022.
46. Khandaker GM, Cousins L, Deakin J, Lennox BR, Yolken R, Jones PB. Inflammation and immunity in schizophrenia: implications for pathophysiology and treatment. Lancet Psychiatry. 2015;2(3):258–70. https://doi.org/10.1016/S2215-0366(14)00122-9.
47. Blednov YA, Benavidez JM, Black M, Ferguson LB, Schoenhard GL, Goate AM, Edenberg HJ, Wetherill L, Hesselbrock V, Foroud T, Harris RA. Peroxisome proliferator-activated receptors α and γ are linked with alcohol consumption in mice and withdrawal and dependence in humans. Alcohol Clin Exp Res. 2015;39(1):136–45. https://doi.org/10.1111/acer.12610.
48. Benedetti E, Cristiano L, Antonosante A, d'Angelo M, D'Angelo B, Selli S, Castelli V, Ippoliti R, Giordano A, Cimini A. PPARs in neurodegenerative and neuroinflammatory pathways. Curr Alzheimer Res. 2018;15(4):336–44. https://doi.org/10.2174/1567205014666170517150037.
49. D'Angelo M, Castelli V, Tupone MG, Catanesi M, Antonosante A, Dominguez-Benot R, Ippoliti R, Cimini AM, Benedetti E. Lifestyle and food habits impact on chronic diseases: roles of PPARs. Int J Mol Sci. 2019;20(21):5422. https://doi.org/10.3390/ijms20215422.
50. Miller BJ, Buckley P, Seabolt W, Mellor A, Kirkpatrick B. Meta-analysis of cytokine alterations in schizophrenia: clinical status and antipsychotic effects. Biol Psychiatry. 2011;70(7):663–71. https://doi.org/10.1016/j.biopsych.2011.04.013.
51. García-Álvarez L, Caso JR, García-Portilla MP, de la Fuente-Tomás L, González-Blanco L, Sáiz Martínez P, Leza JC, Bobes J. Regulation of inflammatory pathways in schizophrenia: a comparative study with bipolar disorder and healthy controls. Eur Psychiatry. 2018;47:50–9. https://doi.org/10.1016/j.eurpsy.2017.09.007.
52. Müller N. Inflammation in schizophrenia: pathogenetic aspects and therapeutic considerations. Schizophr Bull. 2018;44(5):973–82. https://doi.org/10.1093/schbul/sby024.
53. Zeni-Graiff M, Rizzo LB, Mansur RB, Maurya PK, Sethi S, Cunha GR, Asevedo E, Pan P, Zugman A, Yamagata AS, Higuchi C, Bressan RA, Gadelha A, Brietzke E. Peripheral

immuno-inflammatory abnormalities in ultra-high risk of developing psychosis. Schizophr Res. 2016;176(2–3):191–5. https://doi.org/10.1016/j.schres.2016.06.031.
54. Balan I, Beattie MC, O'Buckley TK, Aurelian L, Morrow AL. Endogenous neurosteroid (3α,5α)3-hydroxypregnan-20-one inhibits toll-like-4 receptor activation and pro-inflammatory signaling in macrophages and brain. Sci Rep. 2019;9:1220.
55. Balan I, et al. Neurosteroid allopregnanolone (3α,5α-THP) inhibits inflammatory signals induced by activated MyD88-dependent toll-like receptors. Transl Psychiatry. 2021;11:145.
56. Sullivan CR, Mielnik CA, O'Donovan SM, Funk AJ, Bentea E, DePasquale EA, Alganem K, Wen Z, Haroutunian V, Katsel P, Ramsey AJ, Meller J, McCullumsmith RE. Connectivity analyses of bioenergetic changes in schizophrenia: identification of novel treatments. Mol Neurobiol. 2019;56(6):4492–517. https://doi.org/10.1007/s12035-018-1390-4.
57. De Felice M, Melis M, Aroni S, Muntoni AL, Fanni S, Frau R, Devoto P, Pistis M. The PPARα agonist fenofibrate attenuates disruption of dopamine function in a maternal immune activation rat model of schizophrenia. CNS Neurosci Ther. 2019;25(5):549–61. https://doi.org/10.1111/cns.13087.
58. Lian J, Huang XF, Pai N, Deng C. Ameliorating antipsychotic-induced weight gain by betahistine: mechanisms and clinical implications. Pharmacol Res. 2016;106:51–63. https://doi.org/10.1016/j.phrs.2016.02.011.
59. D'Aniello E, Fellous T, Iannotti FA, Gentile A, Allarà M, Balestrieri F, Gray R, Amodeo P, Vitale RM, Di Marzo V. Identification and characterization of phytocannabinoids as novel dual PPARα/γ agonists by a computational and in vitro experimental approach. Biochim Biophy Acta Gen Subj. 2019;1863(3):586–97. https://doi.org/10.1016/j.bbagen.2019.01.002.
60. Najjar RS, Feresin RG. Plant-based diets in the reduction of body fat: physiological effects and biochemical insights. Nutrients. 2019;11(11):2712. https://doi.org/10.3390/nu11112712.
61. McKhann GM, Knopman DS, Chertkow H, Hyman BT, Jack CR Jr, Kawas CH, Klunk WE, Koroshetz WJ, Manly JJ, Mayeux R, Mohs RC, Morris JC, Rossor MN, Scheltens P, Carrillo MC, Thies B, Weintraub S, Phelps CH. The diagnosis of dementia due to Alzheimer's disease: recommendations from the National Institute on aging-Alzheimer's association workgroups on diagnostic guidelines for Alzheimer's disease. Alzheimers Dement. 2011;7(3):263–9. https://doi.org/10.1016/j.jalz.2011.03.005.
62. Hardy J, Selkoe DJ. The amyloid hypothesis of Alzheimer's disease: progress and problems on the road to therapeutics. Science. 2002;297(5580):353–6. https://doi.org/10.1126/science.1072994.
63. Lannfelt L, Relkin NR, Siemers ER. Amyloid-ß-directed immunotherapy for Alzheimer's disease. J Intern Med. 2014;275(3):284–95. https://doi.org/10.1111/joim.12168.
64. Heneka, M. T., Carson, M. J., El Khoury, J., Landreth, G. E., Brosseron, F., Feinstein, D. L., , . . . Kummer, M. P. (2015). Neuroinflammation in Alzheimer's disease. Lancet Neurol, 14(4), 388–405. doi:https://doi.org/10.1016/S1474-4422(15)70016-5
65. Leng F, Edison P. Neuroinflammation and microglial activation in Alzheimer'sdisease: where do we go from here? Nat Rev Neurol. 2021;17(3):157–72. https://doi.org/10.1038/s41582-020-00435-y.
66. DiSabato DJ, Quan N, Godbout JP. Neuroinflammation: the devil is in the details. J Neurochem. 2016;139(Suppl 2):136–53. https://doi.org/10.1111/jnc.13607.
67. Munhoz CD, García-Bueno B, Madrigal JL, Lepsch LB, Scavone C, Leza JC. Stress-induced neuroinflammation: mechanisms and new pharmacological targets. Braz J of Med Biol Res. 2008;41(12):1037–46. https://doi.org/10.1590/s0100-879x2008001200001.
68. Lyman M, Lloyd DG, Ji X, Vizcaychipi MP, Ma D. Neuroinflammation: the role and consequences. Neurosci Res. 2014;79:1–12. https://doi.org/10.1016/j.neures.2013.10.004.
69. Wójtowicz S, Strosznajder AK, Jeżyna M, Strosznajder JB. The novel role of PPAR alpha in the brain: promising target in therapy of Alzheimer's disease and other neurodegenerative disorders. Neurochem Res. 2020;45(5):972–88. https://doi.org/10.1007/s11064-020-02993-5.
70. Luo R, Su LY, Li G, Yang J, Liu Q, Yang LX, Zhang DF, Zhou H, Xu M, Fan Y, Li J, Yao YG. Activation of PPARA-mediated autophagy reduces Alzheimer'sdisease-like pathology

and cognitive decline in a murine model. Autophagy. 2020;16(1):52–69. https://doi.org/10.1080/15548627.2019.1596488.
71. Nisbett KE, Pinna G. Emerging therapeutic role of PPAR-α in cognition and emotions. Front Pharmacol. 2018;9:998. https://doi.org/10.3389/fphar.2018.00998.
72. Samii A, Nutt JG, Ransom BR. Parkinson's disease. Lancet. 2004;363(9423):1783–93. https://doi.org/10.1016/S0140-6736(04)16305-8.
73. Emre M. Dementia associated with Parkinson's disease. Lancet Neurol. 2003;2(4):229–37. https://doi.org/10.1016/s1474-4422(03)00351-x.
74. McDonald WM, Richard IH, DeLong MR. Prevalence, etiology, and treatment of depression in Parkinson's disease. Biol Psychiatry. 2003;54(3):363–75. https://doi.org/10.1016/s0006-3223(03)00530-4.
75. Schneider RB, Iourinets J, Richard IH. Parkinson's disease psychosis: presentation, diagnosis and management. Neurodegener Dis Manag. 2017;7(6):365–76. https://doi.org/10.2217/nmt-2017-0028.
76. Cacabelos R. Parkinson's disease: from pathogenesis to pharmacogenomics. Int J Mol Sci. 2017;18(3):551. https://doi.org/10.3390/ijms18030551.
77. Rocha EM, De Miranda B, Sanders LH. Alpha-synuclein: pathology, mitochondrial dysfunction and neuroinflammation in Parkinson's disease. Neurobiol Dis. 2018;109(Pt B):249–57. https://doi.org/10.1016/j.nbd.2017.04.004.
78. Marogianni C, Sokratous M, Dardiotis E, Hadjigeorgiou GM, Bogdanos D, Xiromerisiou G. Neurodegeneration and inflammation-an interesting interplay in Parkinson's disease. Int J Mol Sci. 2020;21(22):8421. https://doi.org/10.3390/ijms21228421.
79. Singer TP, Ramsay RR. Mechanism of the neurotoxicity of MPTP. An update. FEBS Lett. 1990;274(1–2):1–8. https://doi.org/10.1016/0014-5793(90)81315-f.
80. Simon DK, Simuni T, Elm J, Clark-Matott J, Graebner AK, Baker L, Dunlop SR, Emborg M, Kamp C, Morgan JC, Ross GW, Sharma S, Ravina B, Investigators NINDSNET-PD. Peripheral biomarkers of Parkinson's disease progression and pioglitazone effects. J Parkinsons Dis. 2015;5(4):731–6. https://doi.org/10.3233/JPD-150666.
81. Pisanu A, Lecca D, Mulas G, Wardas J, Simbula G, Spiga S, Carta AR. Dynamic changes in pro- and anti-inflammatory cytokines in microglia after PPAR-γ agonist neuroprotective treatment in the MPTPp mouse model of progressive Parkinson's disease. Neurobiol Dis. 2014;71:280–91. https://doi.org/10.1016/j.nbd.2014.08.011.
82. Lee Y, Cho JH, Lee S, Lee W, Chang SC, Chung HY, Moon HR, Lee J. Neuroprotective effects of MHY908, a PPAR α/γ dual agonist, in a MPTP-induced Parkinson's disease model. Brain Res. 2019;1704:47–58. https://doi.org/10.1016/j.brainres.2018.09.036.
83. Bordet R, Ouk T, Petrault O, Gelé P, Gautier S, Laprais M, Deplanque D, Duriez P, Staels B, Fruchart JC, Bastide M. PPAR: a new pharmacological target for neuroprotection in stroke and neurodegenerative diseases. Biochem Soc Trans. 2006;34(Pt 6):1341–6. https://doi.org/10.1042/BST0341341.
84. Kreisler A, Duhamel A, Vanbesien-Mailliot C, Destée A, Bordet R. Differing short-term neuroprotective effects of the fibrates fenofibrate and bezafibrate in MPTP and 6-OHDA experimental models of Parkinson's disease. Behav Pharmacol. 2010;21(3):194–205. https://doi.org/10.1097/FBP.0b013e32833a5c81.
85. Zhao C, Zhu J, Liu X, Pu C, Lai Y, Chen L, Yu X, Hong N. Structural and functional brain abnormalities in schizophrenia: a cross-sectional study at different stages of the disease. Prog Neuro-Psychopharmacol Biol Psychiatry. 2018;83:27–32. https://doi.org/10.1016/j.pnpbp.2017.12.017.
86. Torres US, Portela-Oliveira E, Borgwardt S, Busatto GF. Structural brain changes associated with antipsychotic treatment in schizophrenia as revealed by voxel-based morphometric MRI: an activation likelihood estimation meta-analysis. BMC Psychiatry. 2013;13:342. https://doi.org/10.1186/1471-244X-13-342.
87. Lewine R, Hart M. Schizophrenia spectrum and other psychotic disorders. Handb Clin Neurol. 2020;175:315–33. https://doi.org/10.1016/B978-0-444-64123-6.00022-9.

88. Millan MJ. An epigenetic framework for neurodevelopmental disorders: from pathogenesis to potential therapy. Neuropharmacology. 2013;68:2–82. https://doi.org/10.1016/j.neuropharm.2012.11.015.
89. Millan MJ, Andrieux A, Bartzokis G, Cadenhead K, Dazzan P, Fusar-Poli P, Gallinat J, Giedd J, Grayson DR, Heinrichs M, Kahn R, Krebs MO, Leboyer M, Lewis D, Marin O, Marin P, Meyer-Lindenberg A, McGorry P, McGuire P, Owen MJ, et al. Altering the course of schizophrenia: progress and perspectives. Nat Rev Drug Discov. 2016;15(7):485–515. https://doi.org/10.1038/nrd.2016.28.
90. García-Bueno B, Bioque M, Mac-Dowell KS, Barcones MF, Martínez-Cengotitabengoa M, Pina-Camacho L, et al. Pro−/anti-inflammatory dysregulation in patients with first episode of psychosis: toward an integrative inflammatory hypothesis of schizophrenia. Schizophr Bull. 2014;40(2):376–87. https://doi.org/10.1093/schbul/sbt001.
91. Na KS, Kim YK. Monocytic, Th1 and th2 cytokine alterations in the pathophysiology of schizophrenia. Neuropsychobiology. 2007;56(2–3):55–63. https://doi.org/10.1159/000111535.
92. Maes M. Cytokines in schizophrenia. Biol Psychiatry. 1997;42(4):308–9. https://doi.org/10.1016/S0006-3223(97)00240-0.
93. Fond G, Godin O, Boyer L, Berna F, Andrianarisoa M, Coulon N, Brunel L, Bulzacka E, Aouizerate B, Capdevielle D, Chereau I, D'Amato T, Dubertret C, Dubreucq J, Faget C, Leignier S, Lançon C, Mallet J, Misdrahi D, Passerieux C, et al. Chronic low-grade peripheral inflammation is associated with ultra resistant schizophrenia. Results from the FACE-SZ cohort. Eur Arch Psychiatry Clin Neurosci. 2019;269(8):985–92. https://doi.org/10.1007/s00406-018-0908-0.
94. Leboyer M, Godin O, Terro E, Boukouaci W, Lu CL, Andre M, et al. Immune signatures of treatment-resistant schizophrenia: a fondamental academic centers of expertise for schizophrenia (FACE-SZ) study. Schizophr Bull Open. 2021;2(1):sgab012. https://doi.org/10.1093/schizbullopen/sgab012.
95. Labonté C, Zhand N, Park A, Harvey PD. Complete blood count inflammatory markers in treatment-resistant schizophrenia: evidence of association between treatment responsiveness and levels of inflammation. Psychiatry Res. 2022;308:114382. https://doi.org/10.1016/j.psychres.2021.114382.
96. Fineberg AM, Ellman LM. Inflammatory cytokines and neurological and neurocognitive alterations in the course of schizophrenia. Biol Psychiatry. 2013;73(10):951–66. https://doi.org/10.1016/j.biopsych.2013.01.001.
97. Bora E. Peripheral inflammatory and neurotrophic biomarkers of cognitive impairment in schizophrenia: a meta-analysis. Psychol Med. 2019;49(12):1971–9. https://doi.org/10.1017/S0033291719001685.
98. Murphy CE, Walker AK, O'Donnell M, Galletly C, Lloyd AR, Liu D, Weickert CS, Weickert TW. Peripheral NF-κB dysregulation in people with schizophrenia drives inflammation: putative anti-inflammatory functions of NF-κB kinases. Transl Psychiatry. 2022;12(1):21. https://doi.org/10.1038/s41398-021-01764-2.
99. Riazi K, Galic MA, Kentner AC, Reid AY, Sharkey KA, Pittman QJ. Microglia-dependent alteration of glutamatergic synaptic transmission and plasticity in the hippocampus during peripheral inflammation. J Neurosci Off J Soc Neurosci. 2015;35(12):4942–52. https://doi.org/10.1523/JNEUROSCI.4485-14.2015.
100. Barron H, Hafizi S, Andreazza AC, Mizrahi R. Neuroinflammation and oxidative stress in psychosis and psychosis risk. Int J Mol Sci. 2017;18(3):651. https://doi.org/10.3390/ijms18030651.
101. Olmos G, Lladó J. Tumor necrosis factor alpha: a link between neuroinflammation and excitotoxicity. Mediat Inflamm. 2014;2014:861231. https://doi.org/10.1155/2014/861231.
102. Cai H, Cao T, Zhou X, Yao JK. Neurosteroids in schizophrenia: pathogenic and therapeutic implications. Front Psych. 2018;9:73. https://doi.org/10.3389/fpsyt.2018.00073.

103. Locci A, Pinna G. Neurosteroid biosynthesis down-regulation and changes in $GABA_A$ receptor subunit composition: a biomarker axis in stress-induced cognitive and emotional impairment. Br J Pharmacol. 2017;174(19):3226–41. https://doi.org/10.1111/bph.13843.
104. Javitt DC. Glutamate as a therapeutic target in psychiatric disorders. Mol Psychiatry. 2004;9 (11):984–79. https://doi.org/10.1038/sj.mp.4001551.
105. Vyklicky V, Smejkalova T, Krausova B, Balik A, Korinek M, Borovska J, Horak M, Chvojkova M, Kleteckova L, Vales K, Cerny J, Nekardova M, Chodounska H, Kudova E, Vyklicky L. Preferential inhibition of tonically over phasically activated NMDA receptors by pregnane derivatives. J Neurosci Off J Soc Neurosci. 2016;36(7):2161–75. https://doi.org/10.1523/JNEUROSCI.3181-15.2016.
106. Howes OD, McCutcheon R, Agid O, de Bartolomeis A, van Beveren NJ, Birnbaum ML, Bloomfield MA, Bressan RA, Buchanan RW, Carpenter WT, Castle DJ, Citrome L, Daskalakis ZJ, Davidson M, Drake RJ, Dursun S, Ebdrup BH, Elkis H, Falkai P, Fleischacker WW, et al. Treatment-resistant schizophrenia: treatment response and resistance in psychosis (TRRIP) working group consensus guidelines on diagnosis and terminology. Am J Psychiatry. 2017;174(3):216–29. https://doi.org/10.1176/appi.ajp.2016.16050503.
107. Kennedy JL, Altar CA, Taylor DL, Degtiar I, Hornberger JC. The social and economic burden of treatment-resistant schizophrenia: a systematic literature review. Int Clin Psychopharmacol. 2014;29(2):63–76.
108. Barnes TR, Drake R, Paton C, Cooper SJ, Deakin B, Ferrier IN, Gregory CJ, Haddad PM, Howes OD, Jones I, Joyce EM, Lewis S, Lingford-Hughes A, MacCabe JH, Owens DC, Patel MX, Sinclair JM, Stone JM, Talbot PS, Upthegrove R, et al. Evidence-based guidelines for the pharmacological treatment of schizophrenia: updated recommendations from the British Association for Psychopharmacology. J Psychopharmacol. 2020;34(1):3–78. https://doi.org/10.1177/0269881119889296.
109. Potkin SG, Kane JM, Correll CU, Lindenmayer JP, Agid O, Marder SR, Olfson M, Howes OD. The neurobiology of treatment-resistant schizophrenia: paths to antipsychotic resistance and a roadmap for future research. NPJ Schizophr. 2020;6(1):1. https://doi.org/10.1038/s41537-019-0090-z.
110. Keller WR, Kum LM, Wehring HJ, Koola MM, Buchanan RW, Kelly DL. A review of anti-inflammatory agents for symptoms of schizophrenia. J Psychopharmacol. 2013;27(4):337–42. https://doi.org/10.1177/0269881112467089.
111. Meltzer HY. Treatment-resistant schizophrenia--the role of clozapine. Curr Med Res Opin. 1997;14(1):1–20. https://doi.org/10.1185/03007999709113338.
112. Siskind D, Siskind V, Kisely S. Clozapine response rates among people with treatment-resistant schizophrenia: data from a systematic review and meta-analysis. Can J Psychiatry. 2017;62(11):772–7. https://doi.org/10.1177/0706743717718167.
113. Fernandes BS, Steiner J, Bernstein HG, Dodd S, Pasco JA, Dean OM, Nardin P, Gonçalves CA, Berk M. C-reactive protein is increased in schizophrenia but is not altered by antipsychotics: meta-analysis and implications. Mol Psychiatry. 2016;21(4):554–64. https://doi.org/10.1038/mp.2015.87.
114. Mondelli V, Ciufolini S, Belvederi Murri M, Bonaccorso S, Di Forti M, Giordano A, Marques TR, Zunszain PA, Morgan C, Murray RM, Pariante CM, Dazzan P. Cortisol and inflammatory biomarkers predict poor treatment response in first episode psychosis. Schizophr Bull. 2015;41 (5):1162–70. https://doi.org/10.1093/schbul/sbv028.
115. Kose M, Pariante CM, Dazzan P, Mondelli V. The role of peripheral inflammation in clinical outcome and brain imaging abnormalities in psychosis: a systematic review. Front Psych. 2021;12:612471. https://doi.org/10.3389/fpsyt.2021.612471.
116. Guang S, Pang N, Deng X, Yang L, He F, Wu L, Chen C, Yin F, Peng J. Synaptopathology involved in autism Spectrum disorder. Front Cell Neurosci. 2018;12:470. https://doi.org/10.3389/fncel.2018.00470.
117. Masini E, Loi E, Vega-Benedetti AF, Carta M, Doneddu G, Fadda R, Zavattari P. An overview of the Main genetic, epigenetic and environmental factors involved in autism Spectrum

disorder focusing on synaptic activity. Int J Mol Sci. 2020;21(21):8290. https://doi.org/10.3390/ijms21218290.
118. Yoon SH, Choi J, Lee WJ, Do JT. Genetic and epigenetic Etiology underlying autism Spectrum disorder. J Clin Med. 2020;9(4):966. https://doi.org/10.3390/jcm9040966.
119. Matta SM, Hill-Yardin EL, Crack PJ. The influence of neuroinflammation in autism Spectrum disorder. Brain Behav Immun. 2019;79:75–90. https://doi.org/10.1016/j.bbi.2019.04.037.
120. Meltzer A, Van de Water J. The role of the immune system in autism Spectrum disorder. *Neuropsychopharmacology: official publication of the American college of*. Neuropsychopharmacology. 2017;42(1):284–98. https://doi.org/10.1038/npp.2016.158.
121. Prata J, Machado AS, von Doellinger O, Almeida MI, Barbosa MA, Coelho R, Santos SG. The contribution of inflammation to autism Spectrum disorders: recent clinical evidence. Methods Mol Biol. 2019;2011:493–510. https://doi.org/10.1007/978-1-4939-9554-7_29.
122. Thom RP, Keary CJ, Palumbo ML, Ravichandran CT, Mullett JE, Hazen EP, Neumeyer AM, McDougle CJ. Beyond the brain: a multi-system inflammatory subtype of autism spectrum disorder. Psychopharmacology. 2019;236(10):3045–61. https://doi.org/10.1007/s00213-019-05280-6.
123. Barone R, Rizzo R, Tabbì G, Malaguarnera M, Frye RE, Bastin J. Nuclear peroxisome proliferator-activated receptors (PPARs) as therapeutic targets of resveratrol for autism Spectrum disorder. Int J Mol Sci. 2019;20(8):1878. https://doi.org/10.3390/ijms20081878.
124. Capano L, Dupuis A, Brian J, Mankad D, Genore L, Hastie Adams R, Smile S, Lui T, Odrobina D, Foster JA, Anagnostou E. A pilot dose finding study of pioglitazone in autistic children. Mol Autism. 2018;9:59. https://doi.org/10.1186/s13229-018-0241-5.
125. Ghaleiha A, Rasa SM, Nikoo M, Farokhnia M, Mohammadi MR, Akhondzadeh S. A pilot double-blind placebo-controlled trial of pioglitazone as adjunctive treatment to risperidone: effects on aberrant behavior in children with autism. Psychiatry Res. 2015;229(1–2):181–7. https://doi.org/10.1016/j.psychres.2015.07.043.
126. Kirsten TB, Casarin RC, Bernardi MM, Felicio LF. Pioglitazone abolishes autistic-like behaviors via the IL-6 pathway. PLoS One. 2018;13(5):e0197060. https://doi.org/10.1371/journal.pone.0197060.
127. Mirza R, Sharma B. Benefits of Fenofibrate in prenatal valproic acid-induced autism spectrum disorder related phenotype in rats. Brain Res Bull. 2019;147:36–46. https://doi.org/10.1016/j.brainresbull.2019.02.003.
128. D'Agostino G, Cristiano C, Lyons DJ, Citraro R, Russo E, Avagliano C, Russo R, Raso GM, Meli R, De Sarro G, Heisler LK, Calignano A. Peroxisome proliferator-activated receptor alpha plays a crucial role in behavioral repetition and cognitive flexibility in mice. Molecular Metab. 2015;4(7):528–36. https://doi.org/10.1016/j.molmet.2015.04.005.
129. Cristiano C, Pirozzi C, Coretti L, Cavaliere G, Lama A, Russo R, Lembo F, Mollica MP, Meli R, Calignano A, Mattace Raso G. Palmitoylethanolamide counteracts autistic-like behaviours in BTBR T+tf/J mice: contribution of central and peripheral mechanisms. Brain Behav Immun. 2018;74:166–75. https://doi.org/10.1016/j.bbi.2018.09.003.
130. Aleshin S, Strokin M, Sergeeva M, Reiser G. Peroxisome proliferator-activated receptor (PPAR)β/δ, a possible nexus of PPARα- and PPARγ-dependent molecular pathways in neurodegenerative diseases: review and novel hypotheses. Neurochem Int. 2013;63(4):322–30. https://doi.org/10.1016/j.neuint.2013.06.012.
131. Song L, Wang H, Wang YJ, Wang JL, Zhu Q, Wu F, Zhang W, Jiang B. Hippocampal PPARα is a novel therapeutic target for depression and mediates the antidepressant actions of fluoxetine in mice. Br J Pharmacol. 2018;175(14):2968–87. https://doi.org/10.1111/bph.14346.
132. Ahmad SF, Nadeem A, Ansari MA, Bakheet SA, Alshammari MA, Attia SM. The PPARδ agonist GW0742 restores neuroimmune function by regulating Tim-3 and Th17/Treg-related signaling in the BTBR autistic mouse model. Neurochem Int. 2018;120:251–61. https://doi.org/10.1016/j.neuint.2018.09.006. American Psychiatric Association. Diagnostic and Statistical Manual of Mental Disorders, 5th edn. American Psychiatric Publishing, Arlington, VA, 2013

133. Dowlati Y, Herrmann N, Swardfager W, Liu H, Sham L, Reim EK, Lanctôt KL. A meta-analysis of cytokines in major depression. Biol Psychiatry. 2010;67(5):446–57. https://doi.org/10.1016/j.biopsych.2009.09.033.
134. Fleshner M, Frank M, Maier SF. Danger signals and Inflammasomes: stress-evoked sterile inflammation in mood disorders. *Neuropsychopharmacology: official publication of the American college of*. Neuropsychopharmacology. 2017;42(1):36–45. https://doi.org/10.1038/npp.2016.125.
135. Maes M, Yirmyia R, Noraberg J, Brene S, Hibbeln J, Perini G, Kubera M, Bob P, Lerer B, Maj M. The inflammatory & neurodegenerative (I&ND) hypothesis of depression: leads for future research and new drug developments in depression. Metab Brain Dis. 2009;24(1):27–53. https://doi.org/10.1007/s11011-008-9118-1.
136. Hong F, Xu P, Zhai Y. The opportunities and challenges of peroxisome proliferator-activated receptors ligands in clinical drug discovery and development. Int J Mol Sci. 2018;19(8):2189. https://doi.org/10.3390/ijms19082189.
137. Kemp DE, Schinagle M, Gao K, Conroy C, Ganocy SJ, Ismail-Beigi F, Calabrese JR. PPAR-γ agonism as a modulator of mood: proof-of-concept for pioglitazone in bipolar depression. CNS Drugs. 2014;28(6):571–81. https://doi.org/10.1007/s40263-014-0158-2.
138. Sepanjnia K, Modabbernia A, Ashrafi M, Modabbernia MJ, Akhondzadeh S. Pioglitazone adjunctive therapy for moderate-to-severe major depressive disorder: randomized double-blind placebo-controlled trial. Neuropsychopharmacology. 2012;37(9):2093–100. https://doi.org/10.1038/npp.2012.58.
139. Sasso O, La Rana G, Vitiello S, Russo R, D'Agostino G, Iacono A, Russo E, Citraro R, Cuzzocrea S, Piazza PV, De Sarro G, Meli R, Calignano A. Palmitoylethanolamide modulates pentobarbital-evoked hypnotic effect in mice: involvement of allopregnanolone biosynthesis. Eur Neuropsychopharmacol. 2010;20(3):195–206. https://doi.org/10.1016/j.euroneuro.2009.09.003.
140. Sasso O, Russo R, Vitiello S, Raso GM, D'Agostino G, Iacono A, La Rana G, Vallée M, Cuzzocrea S, Piazza PV, Meli R, Calignano A. Implication of allopregnanolone in the antinociceptive effect of N-palmitoylethanolamide in acute or persistent pain. Pain. 2012;153(1):33–41. https://doi.org/10.1016/j.pain.2011.08.010.
141. Pinna G. Animal models of PTSD: the socially isolated mouse and the biomarker role of allopregnanolone. Front Behav Neurosci. 2019;13:114. https://doi.org/10.3389/fnbeh.2019.00114. PMID: 31244621; PMCID: PMC6579844
142. Pinna G. Endocannabinoids and precision medicine for mood disorders and suicide. Front Psych. 2021;12:658433. https://doi.org/10.3389/fpsyt.2021.658433. PMID: 34093274; PMCID: PMC8173054
143. Sinyor M, Schaffer A, Levitt A. The sequenced treatment alternatives to relieve depression (STAR*D) trial: a review. Can J Psychiatry. 2010;55(3):126–35. https://doi.org/10.1177/070674371005500303.
144. McIntyre RS, Filteau MJ, Martin L, Patry S, Carvalho A, Cha DS, Barakat M, Miguelez M. Treatment-resistant depression: definitions, review of the evidence, and algorithmic approach. J Affect Disord. 2014;156:1–7. https://doi.org/10.1016/j.jad.2013.10.043.
145. Halaris A, Sohl E, Whitham EA. Treatment-resistant depression revisited: a glimmer of hope. J Pers Med. 2021;11(2):155. https://doi.org/10.3390/jpm11020155.
146. Lauden A, Geishin A, Merzon E, Korobeinikov A, Green I, Golan-Cohen A, Vinker S, Manor I, Weizman A, Magen E. Higher rates of allergies, autoimmune diseases and low-grade inflammation markers in treatment-resistant major depression. Brain Behav Immun Health. 2021;16:100313. https://doi.org/10.1016/j.bbih.2021.100313.
147. Strawbridge R, Hodsoll J, Powell TR, Hotopf M, Hatch SL, Breen G, Cleare AJ. Inflammatory profiles of severe treatment-resistant depression. J Affect Disord. 2019;246:42–51. https://doi.org/10.1016/j.jad.2018.12.037.
148. Soczynska JK, Mansur RB, Brietzke E, Swardfager W, Kennedy SH, Woldeyohannes HO, Powell AM, Manierka MS, McIntyre RS. Novel therapeutic targets in depression: minocycline

as a candidate treatment. Behav Brain Res. 2012;235(2):302–17. https://doi.org/10.1016/j.bbr.2012.07.026.
149. Chamberlain SR, Cavanagh J, de Boer P, Mondelli V, Jones D, Drevets WC, Cowen PJ, Harrison NA, Pointon L, Pariante CM, Bullmore ET. Treatment-resistant depression and peripheral C-reactive protein. Br J Psychiatry J Ment Sci. 2019;214(1):11–9. https://doi.org/10.1192/bjp.2018.66.
150. Kruse JL, Congdon E, Olmstead R, Njau S, Breen EC, Narr KL, Espinoza R, Irwin MR. Inflammation and improvement of depression following electroconvulsive therapy in treatment-resistant depression. J Clin Psychiatry. 2018;79(2):17m11597. https://doi.org/10.4088/JCP.17m11597.
151. Abbasi SH, Hosseini F, Modabbernia A, Ashrafi M, Akhondzadeh S. Effect of celecoxib add-on treatment on symptoms and serum IL-6 concentrations in patients with major depressive disorder: randomized double-blind placebo-controlled study. J Affect Disord. 2012;141(2–3):308–14. https://doi.org/10.1016/j.jad.2012.03.033.
152. Gallagher PJ, Castro V, Fava M, Weilburg JB, Murphy SN, Gainer VS, Churchill SE, Kohane IS, Iosifescu DV, Smoller JW, Perlis RH. Antidepressant response in patients with major depression exposed to NSAIDs: a pharmacovigilance study. Am J Psychiatry. 2012;169(10):1065–72. https://doi.org/10.1176/appi.ajp.2012.11091325.
153. Raison CL, Rutherford RE, Woolwine BJ, Shuo C, Schettler P, Drake DF, Haroon E, Miller AH. A randomized controlled trial of the tumor necrosis factor antagonist infliximab for treatment-resistant depression: the role of baseline inflammatory biomarkers. JAMA Psychiat. 2013;70(1):31–41. https://doi.org/10.1001/2013.jamapsychiatry.4.
154. Yilmaz C, Karali K, Fodelianaki G, Gravanis A, Chavakis T, Charalampopoulos I, Alexaki VI. Neurosteroids as regulators of neuroinflammation. Front Neuroendocrinol. 2019;55:100788. https://doi.org/10.1016/j.yfrne.2019.100788.
155. Pinna G, Dong E, Matsumoto K, Costa E, Guidotti A. In socially isolated mice, the reversal of brain allopregnanolone down-regulation mediates the anti-aggressive action of fluoxetine. Proc Natl Acad Sci. 2003;100:2035–40.
156. Uzunova V, Ceci M, Kohler C, Uzunov DP, Wrynn AS. Region-specific dysregulation of allopregnanolone brain content in the olfactory bulbectomized rat model of depression. Brain Res. 2003;976:1–8.
157. Uzunova V, et al. Chronic antidepressants reverse cerebrocortical allopregnanolone decline in the olfactory-bulbectomized rat. Eur J Pharmacol. 2004;486:31–4.
158. American Psychiatric Association. Diagnostic and statistical manual of mental disorders. 5th ed. Arlington, VA: American Psychiatric Publishing; 2013.
159. Friedman MJ, Bernardy NC. Considering future pharmacotherapy for PTSD. Neurosci Lett. 2017;649:181–5. https://doi.org/10.1016/j.neulet.2016.11.048.
160. Neumeister A, Seidel J, Ragen BJ, Pietrzak RH. Translational evidence for a role of endocannabinoids in the etiology and treatment of posttraumatic stress disorder. Psychoneuroendocrinology. 2015;51:577–84. https://doi.org/10.1016/j.psyneuen.2014.10.012.
161. Michopoulos V, Powers A, Gillespie CF, Ressler KJ, Jovanovic T. Inflammation in fear- and anxiety-based disorders: PTSD, GAD, and beyond. Neuropsychopharmacology. 2017;42(1):254–70. https://doi.org/10.1038/npp.2016.146.
162. Miller MW, Lin AP, Wolf EJ, Miller DR. Oxidative stress, inflammation, and neuroprogression in chronic PTSD. Harv Rev Psychiatry. 2018;26(2):57–69. https://doi.org/10.1097/HRP.0000000000000167.
163. Quinones MM, Gallegos AM, Lin FV, Heffner K. Dysregulation of inflammation, neurobiology, and cognitive function in PTSD: an integrative review. Cogn Affect Behav Neurosci. 2020;20(3):455–80. https://doi.org/10.3758/s13415-020-00782-9.
164. Rasmusson AM, et al. Decreased cerebrospinal fluid Allopregnanolone levels in women with posttraumatic stress disorder. Biol Psychiatry. 2006;60:704–13.

165. Rasmusson AM, King MW, Valovski I, Gregor K, Scioli-Salter E, Pineles SL, Hamouda M, Nillni YI, Anderson GM, Pinna G. Relationships between cerebrospinal fluid GABAergic neurosteroid levels and symptom severity in men with PTSD. Psychoneuroendocrinology. 2019;102:95–104. https://doi.org/10.1016/j.psyneuen.2018.11.027.
166. Rasmusson AM, Pineles SL, Brown KD, Pinna G. A role for deficits in GABAergic neurosteroids and their metabolites with NMDA receptor antagonist activity in the pathophysiology of posttraumatic stress disorder. J Neuroendocrinol. 2022;34(2):e13062. https://doi.org/10.1111/jne.13062. Epub 2021 Dec 28. PMID: 34962690
167. Passos IC, Vasconcelos-Moreno MP, Costa LG, Kunz M, Brietzke E, Quevedo J, Salum G, Magalhães PV, Kapczinski F, Kauer-Sant'Anna M. Inflammatory markers in post-traumatic stress disorder: a systematic review, meta-analysis, and meta-regression. Lancet Psychiatry. 2015;2(11):1002–12. https://doi.org/10.1016/S2215-0366(15)00309-0.
168. Hill MN, Campolongo P, Yehuda R, Patel S. Integrating endocannabinoid Signaling and cannabinoids into the biology and treatment of posttraumatic stress disorder. Neuropsychopharmacology. 2018;43(1):80–102. https://doi.org/10.1038/npp.2017.162.
169. Steenkamp MM, Blessing EM, Galatzer-Levy IR, Hollahan LC, Anderson WT. Marijuana and other cannabinoids as a treatment for posttraumatic stress disorder: a literature review. Depress Anxiety. 2017;34(3):207–16. https://doi.org/10.1002/da.22596.

# Ketogenic Diet and Inflammation: Implications for Mood and Anxiety Disorders

23

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**Abstract**

The ketogenic diet, known as a low-carbohydrate, high-protein, and high-fat diet, drastically restrains the major source of energy for the body, forcing it to burn all excess fat through a process called ketosis—the breaking down of fat into ketone bodies. First suggested as a medical treatment for children suffering from epilepsy, this diet has gained increased popularity as a rapid weight loss strategy. Over the past few years, there have been numerous studies suggesting that the ketogenic diet may provide therapeutic effects for several psychiatric conditions such as mood- and anxiety-related disorders. However, despite significant progress in research, the mechanisms underlying its therapeutic effects remain largely unexplored and are yet to be fully elucidated. This chapter provides an in-depth overview of preclinical and clinical evidence supporting the use of a ketogenic diet in the management of mood and anxiety disorders and discusses its relationship with inflammatory processes and potential mechanisms of actions for its therapeutic effects.

**Keywords**

Anxiety · Bipolar disorder · Inflammation · Ketogenic diet · Major depressive disorder · Mood disorders · Schizophrenia

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## 23.1 Introduction

The ketogenic diet, commonly known as the "keto diet," is one of the many low-carbohydrate regimes, consisting specifically of a high fat intake of up to 55–60% of total daily calories [1]. It is usually composed of a 4:1 ratio of lipid/non-lipid [2]. This form of diet was first proposed by American physician Russell Wilder as a therapeutic agent to treat epilepsy in children [1]. However, its use diminished as modern medicine became more prominent. Nowadays, this diet has gained increasing popularity as a rapid weight loss formula [1].

An individual with a healthy diet normally utilizes carbohydrates as the primary source of energy. However, when an individual opts for the ketogenic diet, his body becomes deprived of carbohydrates, forcing it to enter into a catabolic state, with fat being now the primary source of energy [1]. This causes insulin secretions to reduce and glycogen storage to deplete, leading to gluconeogenesis and ketogenesis [1]. When glucose levels drop, this process is not able to keep up with the body's great need for glucose; hence, ketogenesis begins [1]. Ketogenesis in turn produces ketone bodies, which replace glucose and act as an alternate source of energy for the body. At this stage, insulin secretion is low due to low blood glucose feedback, which reduces the stimulus for fat and glucose storage [1]. In addition, hormonal changes contribute to the increased breakdown of fats into fatty acids as a source of energy for the body [1]. These fatty acids are then broken down into three fat-derived metabolites: acetoacetate (keto acid), beta-hydroxybutyrate (keto acid), and acetone (ketone) [1]. The body is now in a "ketosis state," in which the ketone bodies are now the primary and alternate source of energy as long as the body is deprived of carbohydrates.

Studies conducted over the past few years suggest that the ketogenic diet has several health benefits other than weight loss, one of which includes having anticonvulsant effects, that is, the ability to control seizures [3]. This was mainly explained by the "pH hypothesis" proposed by Bridge and Iob [4], which suggests that the ketogenic diet makes the blood and brain slightly acidic, hence allowing the brain to stop seizures. In fact, this occurs in the absence of carbohydrates, where fats are used as the primary source of energy, which break down into two keto acids and a ketone. In this case, the ketogenic acids decrease the blood's pH, leading to anticonvulsant effects. In addition to treating epilepsy, the ketogenic diet has shown to be an effective strategy for managing type 2 diabetes [5]. Indeed, there seems to be a positive correlation between weight loss and the ketogenic diet due to this regime's ability to reduce appetite and the amount of body fat [6, 7]. In point of fact, it was shown that a low-carbohydrate diet is much more effective in reducing body weight compared to a low-fat diet [8], most likely due to the presence of ketone bodies coming from excessive fatty acid metabolism. Additionally, the ability of ketone bodies to decrease glucose metabolism and reverse insulin resistance was shown to be associated with low hemoglobin A1c (Hb1ac) levels, which in turn has proved to be beneficial in treating type 2 diabetes [5]. Furthermore, preclinical experiments have shown that ketone bodies significantly increase cardiac efficiency by providing oxidizing ketone bodies as a fuel source for the heart, suggesting that the ketogenic

diet may be beneficial for individuals with cardiovascular complications [9], which are hallmarks of type 2 diabetes [10]. The ketogenic diet is also able to lower triglyceride and increase HDL levels in humans, whose effects play a role in reducing the risk of heart disease [11]. Finally, the ketogenic diet has proved to be effective in helping treat cancer due to its ability to reduce insulin levels and cellular proliferation, hence leading to reduced blood glucose level and fuel availability for cancer cells [12].

The ketogenic diet has recently been mentioned in several studies for its beneficial effects on mood and anxiety disorders. The asset of this diet is that it can replace pharmacological treatments used to treat psychiatric disorders, therefore decreasing the occurrence of potential unwanted side effects while being financially advantageous [13]. In addition, this diet can reverse mitochondrial dysfunction and decrease reactive oxygen species (ROS) production, increase energy production, produce antioxidant characteristics, reduce inflammation and oxidative stress, and limit apoptosis, all of which can have beneficial effects in mood and anxiety disorders [14]. However, despite significant progress in research, the beneficial effects of the ketogenic diet in mood and anxiety disorders along with its underlying mechanisms of actions are poorly understood. In this book chapter, the focus will be on the potential therapeutic effect of the ketogenic diet in neuropsychiatric conditions and the role of inflammation in its underlying mechanisms of action. This chapter first starts by describing the symptoms and underlying neurobiological mechanisms of mood and anxiety disorders and their link with inflammation and finally provide an overview of preclinical and clinical studies highlighting the role of the ketogenic diet in inflammation and in the management of these disorders.

## 23.2 Mood and Anxiety Disorders

### 23.2.1 Symptoms and Underlying Neurobiological Mechanisms of Mood Disorders

Mood disorders are one of the biggest issues today, being considered as a leading cause for disabilities and death [15]. Among them, major depressive disorder (MDD) is a debilitating disorder characterized by a variety of emotional and psychological problems [16]. Approximately 10% of the population will experience at least one episode of depressive disorder once in their lifetime [17]. According to the Diagnostic and Statistical Manual of Mental Disorders (DSM-5), symptoms of MDD include low mood, loss of interest or pleasure, significant weight loss or gain, insomnia or hypersomnia, psychomotor retardation or agitation, loss of energy, feelings of worthlessness, difficulty in thinking or concentrating, and suicidal attempts [18]. An individual is diagnosed with MDD if he has at least five of these symptoms during the same 2-week period [18]. Bipolar disorder is another chronic and recurrent mood disorder that affects at least 1% of the population, leading to increased cognitive and functional impairments [19]. It is characterized by huge mood swings ranging from depression to mania or hypomania [20]. According to the

DSM-5, symptoms of bipolar disorder include inflated self-esteem, diminished need for sleep, pressured need to keep talking, racing thoughts, increased in goal-directed activity, engaging in activities with painful consequences, and distracted attention [21]. Bipolar disorder was also shown to be associated with increased risk for cardiovascular diseases [22] and anxiety disorders [23]. Last but not least, schizophrenia is a mood disorder characterized by positive symptoms, which include delusion and hallucination; negative symptoms, which include a lack of motivation and social withdrawal; and cognitive symptoms, which include reduced attention and altered speech [24, 25]. Globally, this disease has a low prevalence (<1%) but imposes a severe burden to the affected individual and their caregivers [26].

There is mounting evidence showing that genetic factors play a significant role in mood disorders [27]. A meta-analysis of twin studies estimates that genetic factors account for approximately 37% in the heritability of MDD [28]. In addition, genome-wide association studies (GWAS) show a genetic overlap between schizophrenia, bipolar disorder, and MDD due to the merging of functional pathways [29]. GWAS have also shown that the ODZ4 gene, which is involved in signaling and neuronal path finding, and the ankyrin 3 (ANK3) gene, which is involved in localization of sodium channels, are involved in bipolar disorder [30]. Bipolar disorder is also associated with variation in the calcium voltage-gated channel subunit alpha 1C (CACNA1C) gene [31], which has been shown to correlate with cognitive disturbances [32]. In addition, epigenetic factors such as hypomethylation of the catechol-O-methyltransferase (COMT) gene were demonstrated to play a significant role in the pathophysiology of bipolar disease and schizophrenia [33]. Similarly, polymorphisms in genes encoding growth factors such as the brain-derived neurotrophic factor, fibroblast growth factor, insulin-like growth factor, and vascular endothelial growth factor have been shown to be associated with the pathophysiology of MDD [16]. Susceptibility to mood disorders may also be influenced by epistatic interactions between genes, such as the genes encoding for the arginine vasopressin receptor 1B (AVPR1B) and the corticotropin-releasing hormone receptor 1 (CRHR1) [34]. These findings indicate that mood disorders can be viewed as multifactorial disorders, resulting not only from polymorphisms in certain susceptibility gene but also from the interaction between genetic and environmental factors.

Neurotransmitters, including serotonin, dopamine, and norepinephrine, have also been shown to play a major role in the development of mood disorders. For instance, reduced brain serotonin levels and polymorphism of the serotonin transporter gene were shown to be associated with MDD [35]. In addition, a meta-analysis of monoamine depletion studies showed that reduced level of serotonin, dopamine, or norepinephrine was associated with decreased mood in individuals with a family history of MDD and in drug-free patients with MDD in remission [36]. There is also accumulating evidence indicating that schizophrenia is associated with increased dopamine transmission particularly in mesolimbic areas [37], and dopamine receptor blockade through antipsychotics has been the cornerstone of schizophrenia therapy for several decades [38]. The neurotransmitter glutamate has shown to play an important role in the pathophysiology of MDD inasmuch as blockade of the

glutamate N-methyl-D-aspartate (NMDA) receptor exerts rapid and robust antidepressant effects even in treatment-resistant individuals [39]. Bipolar disorder is also positively correlated with excessive sympathetic nervous system activity as evidence indicates that norepinephrine is abnormally elevated in individuals with this disorder [40]. It is also noteworthy to mention that glial changes in brain regions such as the anterior cingulate cortex, prefrontal cortex, orbitofrontal cortex, and amygdala have been demonstrated in individuals with mood disorders, suggesting a link with immune and inflammatory responses [41–43]. In addition, MDD was shown to be associated with decreased responsiveness to glucocorticoids as a result of increased level of inflammatory cytokines [44]. Finally, there is mounting evidence illustrating a relationship between mood disorders and the occurrence of gray matter abnormalities and structural changes in several brain structures in particular those within the frontal-subcortical circuit and the mesolimbic reward circuit [45, 46].

### 23.2.2 Anxiety Disorders: Link with Inflammation

Listed as a serious mental health disorder, anxiety can be subdivided into numerous diagnostic categories including social phobias, post-traumatic stress disorder (PTSD), panic disorders (PD), obsessive-compulsive disorder (OCD), and generalized anxiety disorder (GAD) [47]. Anxiety could be induced either by genetic or dietary contributions; by psychological inputs in early childhood, for example, living with overprotective parents; by social inclusions and environmental factors by means of social pressure such as at school or at work; or by a stressful life event such as the death of a close relative [48, 49]. Some of the biological and dietary factors that are either directly or indirectly associated with stress- and anxiety-like behaviors include the neurotransmitters glutamate, norepinephrine, and GABA [50–52] and dietary intake of magnesium [53]. Inflammation is another main biological factor that is directly linked to anxiety-like disorder [54]. Indeed, exposure to anxiety-provoking stimuli leads to the activation of the hypothalamic-pituitary-adrenal (HPA) axis and the immune system and to the increased release of pro-inflammatory cytokines [55, 56]. Inflammatory cytokines are associated with high levels of oxidative stress as well as the delivery of ROS and reactive nitrogen species (RNS), which in turn alter the cerebral neurocircuits of anxiety by affecting brain regions involved in emotional behaviors such as the amygdala, insula, and prefrontal cortex [57]. In a more profound manner, it was shown that individuals suffering from PTSD show high levels of pro-inflammatory cytokines interleukin-1 beta (IL-1β), IL-6, tumor necrosis alpha (TNF-α), and interferon-gamma (IFN-γ) in the serum and/or cerebrospinal fluid [58, 59] and elevated activity of NF $\kappa$ B [60] and C-reactive protein (CRP) [59]. PTSD was also shown to be associated with changes in immune gene transcription and differential methylation level of genes involved in immune and inflammatory functions [61, 62]. Other anxiogenic factors include changes in sleep rhythms such as loss of sleep hours or interruptions in usual sleep cycles, which can accelerate the production of IL-6 and CRP, creating further inflammatory reactions that promote anxiety [63, 64]. Finally, numerous studies

point to the fact that anxiety disorders are characterized by structural and functional changes in certain brain regions, such as the amygdala, hippocampus, medial prefrontal cortex (mPFC), and anterior cingulate cortex (ACC), most of which are known to play important roles in inflammatory processes [57]. For instance, increases in IL-6 levels—either through disruption in sleep or stress—were shown to play a major role in linking the functions of both the amygdala and the mPFC in promoting anxiety [65]. In addition, evidence indicates that anxiety disorders are associated with smaller hippocampal [66, 67] and amygdala [68, 69] volume, as well as reduced anterior cingulate gray matter density [69]. Anxiety disorders were also shown to be associated with increased activation of the insula and decreased connectivity between the anterior insula, amygdala, and anterior cingulate cortex [70].

Taken together, inflammation has effectively proven to play critical roles in inducing anxiety- and fear-like behaviors and to induce structural and/or functional changes in several brain regions. However, it is important to note that further studies are needed to better understand the relationship between inflammation and anxiety and the underlying mechanisms of action.

## 23.3 Ketogenic Diet and Inflammation

There is mounting evidence suggesting that ketone bodies obtained from the ketogenic diet exert neuroprotective and anti-inflammatory properties. By binding to hydroxy-carboxylic acid receptor 2 (HCA2) expressed on immune cells such as microglia, dendritic cells, and macrophages, β-hydroxybutyrate (BHB)—one of the main ketone bodies detected after supply of a ketogenic diet—can directly influence neuroinflammatory mechanisms [71, 72]. Activation of these receptors by BHB triggers the release of a neuroprotective subset of macrophages leading to dampened inflammation [73, 74]. The ketogenic diet has also been shown to influence neuroinflammatory processes through its inhibitory action on nuclear factor kappa-light-chain-enhancer of activated B cells (NF-kB) [74, 75]. It exerts neuroprotective and anti-inflammatory actions through the activation of microglial cells [76] and its inhibitory effect on the activity of pro-inflammatory cytokines including IL-1β, IL-6, and TNF-α [77]. Preclinical evidence also indicates that the ketogenic diet may exert anti-inflammatory effects through BHB-mediated inhibition of the NLRP3 inflammasome, which controls the release of pro-inflammatory cytokines [78]. This diet was also associated with reduced inflammation through the activation of peroxisome proliferator-activated receptor gamma (PPARγ) [79]. Findings from preclinical experiments correlate with those in clinical settings inasmuch as consumption of a low-carbohydrate diet by overweight individuals with atherogenic dyslipidemia resulted in marked decreases in inflammatory and immune markers including TNF-α, IL-6, IL-8, monocyte chemoattractant protein-1 (MCP-1), and E-selectin [80].

Because of the strong evidence supporting an anti-inflammatory property of ketogenic diet, it is not surprising that many studies have investigated the potential

use of this diet for the treatment of neurodegenerative and psychiatric conditions where inflammatory processes are dysregulated [81]. For instance, the ketogenic diet was shown to be effective in reversing the increased expression of inflammatory cytokines and the production of oxidative stress in a murine model of multiple sclerosis [82]. In a mouse model of Parkinson's disease based on treatments with 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP), the ketogenic diet was shown to exert anti-inflammatory actions by inhibiting the activation of microglia and the level of pro-inflammatory cytokine in the substantia nigra [76]. These molecular changes were associated with a significant alleviation of motor dysfunctions, suggesting a potential therapeutic effect of the ketogenic diet in Parkinson's disease [76]. The ketogenic diet has also shown great promise in treating Alzheimer's disease due to its ability to reduce the expression of inflammatory and apoptotic mediators [74] and in treating conditions involving inflammatory pain such as allodynia [83]. Since inflammatory processes play a key role in the underlying pathophysiology of psychiatric disorders such as anxiety [84], MDD [85], bipolar disorder [86], and schizophrenia [87], it is not surprising that several studies have investigated the therapeutic potential of the ketogenic diet in these disorders.

## 23.4 Ketogenic Diet in Mood Disorders

### 23.4.1 Evidence from Animal Studies

Despite significant improvements in both pharmacological and non-pharmacological treatments, mood disorders, including schizophrenia, MDD, and bipolar disorder, remain at the top of the mood behavior therapeutics agenda displaying a high treatment resistance. Therefore, several studies to date hypothesized nutritional intervention as a promising treatment strategy for mood behaviors. In order to validate those hypotheses, researchers based their studies on animal models of depression, mainly mice and rats, induced either by corticosterone injections [88] or by the chronic social defeat stress model [89, 90].

Several recent studies suggest that a sustained ketogenic diet is of novel therapeutic importance in animal models of schizophrenia [91–93]. For instance, according to Sarnyai and colleagues [91], the ketogenic diet resulted in a complete restoration of normal behavior phenotype in mice models of schizophrenia. In fact, while converging evidence classify brain bioenergetic defects as a cause of schizophrenia and other psychotic disorders [94, 95], the ketogenic diet normalizes brain energy metabolism by circumventing glycolysis and relying on ketosis in order to restore normal brain glucose metabolism and mitochondrial function. Furthermore, by allowing the body to rely on ketone bodies as alternative energy substrates, the ketogenic diet has proven to change the GABA/glutamate ratio in favor of GABA in a way to compensate the low GABA levels displayed in schizophrenic mice by suppressing its catabolism and stimulating its synthesis [91, 92].

Moreover, there is a wide consensus in the literature that the ketogenic diet has similar beneficial effects as conventional antidepressant drugs in the force choice

model of depression in rats [96–98]. In this context, Murphy and colleagues [97] used the Porsolt model—also known as the forced swim test (FST)—to assess the time spent immobile by rats, which indicates behavioral despair, a characteristic of depression. For instance, rats fed with a ketogenic diet and rats treated with antidepressant drugs spent on average less time immobile compared to rats fed with a standard control diet [96, 97]. Since rats on the ketogenic diet are less likely to exhibit behavioral despair, the results suggest that the ketogenic diet has efficient antidepressant properties and in turn could be useful in the treatment of depression. However, despite the aforementioned importance of ketosis in schizophrenia treatment [91–93], a plausible interesting finding of this research is that behavioral change in rats fed with a ketogenic diet was ketosis independent [97]. A likely mechanism mediating the antidepressant effect of the ketogenic diet is the restoration of microglial activation and neuronal excitability in the lateral habenula, a region involved in negative reward processing [99]. There is clearly a need for future research to cautiously examine the importance of ketosis and other potential factors in the antidepressant effect of the ketogenic diet.

Furthermore, evidence suggests that gestational ketogenic diet reduces the susceptibility of mice to depression [100]. In fact, mice prenatally exposed to a ketogenic diet exhibit elevated physical activity in both the FST and the exercise wheel test (EWT) compared to mice exposed to a standard diet prenatally [100]. The following could be explained by neuro-anatomical differences evidenced by magnetic resonance imaging, including a hypothalamic and corpus callosum reduction along with cortical and cerebellar volumetric enlargement [100]. These findings indicate that a prenatal ketogenic diet confers an antidepressant effect even with a postnatal standard diet by inducing neuro-anatomical changes. In addition, while the aforementioned studies neglected gender-based differences [96, 97], this study investigated gender as a covariate of physical activity since female rats fed with a ketogenic diet displayed a significant higher number of rotations on the EWT compared to males [100]. However, all other variables of EWT and FST were gender independent [100].

Difficulties arise, however, when an attempt is made to solely examine the effect of ketogenic diet on animal models of bipolar disorder. In fact, while several clinical approaches investigated the interplay between bipolar disorder in humans and ketogenic diet, no study to date focused on such correlation among animals due to the limited number of suitable experimental models of bipolar disorder available for behavioral and histological analysis [101]. In addition, animal models of bipolar disorder fail to capture all pathophysiological aspects of the disease; only few selected symptoms are displayed, leading to a reduced validity of the model [102].

Taken together, the nutritional intervention of the ketogenic diet is of therapeutic importance in terms of mood disorders in animal models. Although this diet has been shown to exert therapeutic effects in MDD, bipolar disorder, and schizophrenia, several inconsistencies exist in the literature. In addition, many of the studies investigating the therapeutic properties of the ketogenic diet have neglected gender-based differences between male and female rats. For this matter, further studies are required to better examine the therapeutic effect of the ketogenic diet in

mood disorders. At last, despite its efficiency in treating mood disorders, more evidence is needed to assess the long-term safety and tolerability of this diet in order to avoid potential side effects.

### 23.4.2 Evidence from Human Studies

There are converging lines of evidence showing that a ketogenic diet shows beneficial effects in the treatment of mood disorders in humans similar to that observed with traditional medications [2, 103–105]. A study conducted by Campbell and Campbell [106] with bipolar disorder patients showed that the majority of patients (~85%) following a ketogenic diet reported a positive effect on mood stabilization. In another clinical study conducted by Phelps and colleagues [103] on two women diagnosed with bipolar disorder, prolonged ketosis (2 or 3 years) achieved through a ketogenic diet resulted in significant improvement in mental health that exceeded that achieved with medication and led to no signs of adverse effects. On the other hand, a study done with one individual with bipolar disorder showed no positive result with the ketogenic diet keeping in mind ketosis was not achieved [107]. Altogether, these findings indicate that a state of ketosis is necessary for the positive effect and impact of the ketogenic diet on bipolar disorder. Taking this concept another step forward, ketosis is postulated to decrease the level of sodium and calcium intracellularly therefore acting as a mood stabilizer [2, 103, 107, 108].

When it comes to schizophrenia, the ketogenic diet can be considered to be an efficient treatment and can even reverse some of the persistent symptoms of this disease [109]. In another case study, a 33-year-old man diagnosed with MDD and schizoaffective disorder was prescribed with several medications including lamotrigine and lorazepam and then started a ketogenic diet for 3 weeks [110]. Two observations were identified; first, there was a significant reduction on body weight (~15 lb), and second, there was a noticeable improvement in mood and a decrease in schizophrenia-related symptoms including hallucinations [110]. Similar effects were observed in another 31-year-old female patient diagnosed with schizoaffective disorder and prescribed drugs [110]. However, in the latter case, withdrawal from the ketogenic diet led to a severe symptom relapse, and after being back on the diet, it was not before a short fasting period that the symptoms started to disappear [110]. While these findings indicate that the ketogenic diet can successfully be used for the management of schizophrenia, a short fasting is required for this diet to continue exerting its effects if it is discontinued [110]. Interestingly, the medications used by the 31-year-old female patient led to a significant symptom improvement until ketosis was activated, suggesting that the latter can be an important factor in the therapeutic effects of drugs prescribed for schizophrenia [110]. In another study, Kraft and Westman [111] report the case of a female in her 70s with a diagnosis of schizophrenia since the age of 17 and long-standing severe symptoms of paranoia, disorganized speech, and hallucinations. Her doctor suggested a ketogenic diet as she suffered from obesity along with depression [111]. After 7 days of following a ketogenic diet, she already showed a significant amelioration in her

symptoms, and after 19 days, she reported having no more signs of hallucinations [111]. Altogether, these findings demonstrate a strong potential for the ketogenic diet in managing symptoms of schizophrenia.

Moreover, there is strong evidence in humans showing that depressive symptoms can be treated with a ketogenic diet rather than a conventional pharmacological routine [14]. In a study by Cox and colleagues [112], a 12-week ketogenic diet regimen led to decreased glucose levels and fewer symptoms of depression as per the PHQ-9 scale in a patient with MDD and type 2 diabetes. As mentioned previously [110], symptoms of depression are often comorbid with other disorders like bipolar disorder or schizophrenia, and significant mood amelioration was observed following a ketogenic diet, suggesting that this diet increases the quality of life and mental well-being of individuals with mood disorders [13].

Overall, the ketogenic diet is an alternative strategy that is being used and recommended in many cases to treat mood disorders such as bipolar disorder, MDD, and schizophrenia. It can cross out and minimize side effects of medicinal drugs while easing the financial burden to the affected individual. It has been tested and proved that it can reverse the mitochondria dysfunction, which in turn reduces the symptoms of mood behavior [14]. Also, as observed in clinical cases, patients who are suffering from these three major disorders all demonstrate improvement in results and less symptomatology after starting the ketogenic diet as long as ketosis has started. However, some limitations could be noted; for instance, during the online experiments by Campbell and Campbell [106], reports could have been biased and inaccurate, diminishing the validity of the results. Clearly more clinical research needs to be done to better understand the efficiency and mechanisms of action of the ketogenic diet in mood disorders in humans.

## 23.5 Ketogenic Diet in Anxiety Disorders

Over the past few years, there has been an increased interest in using the ketogenic diet in treating anxiety disorder. Individuals with anxiety disorders usually have frequent and persistent fear when dealing with everyday life situations [113]. Anxiety disorders have become more prevalent, possibly due to increased environmental stress and misuse of social media [114, 115]. Anxiety disorders are typically treated with pharmacotherapy and psychotherapy [116], but nutrition seems to have a major impact on these disorders [117].

Numerous studies on experimental animals have investigated the effects of the ketogenic diet on anxiety. Włodarczyk and colleagues [118] and Sussman and colleagues [100] argue that several neurotransmitters are involved in the etiology of anxiety, including serotonin, norepinephrine, glutamate, and GABA. The effects of GABA on anxiety disorders were extensively studied in several preclinical and clinical studies. Through its inhibitory action on neural activity, GABA can regulate anxiety by preventing excessive neuronal excitability [119], and its receptors are often key targets of anxiolytics [52]. As far as the ketogenic diet is concerned, a study conducted by Calderón and colleagues [120] showed that rodents fed with a

ketogenic diet had a significantly higher urine level of GABA compared to rodents fed with a normal diet, suggesting that this diet might be beneficial in reducing anxiety through its action on GABA. Similarly, mice prenatally exposed to a ketogenic diet were shown to travel shorter distances and to spend more time in the center in an open-field test, indicating that this diet may have anxiolytic effects [100]. In addition, results show that male mice visited the center of the open-field apparatus more frequently than female mice regardless of their diet, suggesting that both gender and prenatal diet have an impact on anxiety-related behaviors [100]. The anxiolytic effect of the low-protein intake during gestation is likely due to reduced extracellular release of dopamine in the brain as previously suggested [121, 122]. In another study, chronic administration of ketone supplements to a standard diet was shown to reduce anxiety-like behaviors in rats as assessed by the elevated plus maze test [123]. Concentrations of BHB were significantly elevated, implying the occurrence of ketosis [123]. The anxiolytic properties of KD could also be assessed by investigating its effect on sleep insomuch as individuals with anxiety have reduced sleep quality [124]. In a study conducted by Hallböök and colleagues [125] on 18 children with epilepsy, sleep quality was significantly improved, and the rapid eye movement (REM) was increased following a ketogenic diet for a period of 3 months. In accordance with these findings, a 3-month ketogenic diet therapy significantly improved the sleep anxiety of children with epilepsy, suggesting that this diet may have positive effects on the overall sleep quality of mental well-being of the individual [126].

Taken together, the ketogenic diet can have significant effects on certain neurotransmitter systems and can influence anxiety-related behaviors in a gender-specific manner. However, more studies should be conducted to better understand the anxiolytic effect of the ketogenic diet prior to recommendation for therapeutic purposes over traditional anxiolytic drugs.

## 23.6 Conclusion

Overall, the ketogenic diet has gained increased global popularity not only as a weight loss strategy but also as a therapeutic modality for mood and anxiety disorders. As presented in this chapter, the beneficial effects of the ketogenic diet in mood and/or anxiety disorders may be attributed to a wide variety of mechanisms including the generation of ketone bodies [110, 123], the facilitation of GABAergic transmission [120], the reduction in dopamine release [121, 122], the restoration of microglial activation and neuronal excitability in the lateral habenula [99], the induction of neuro-anatomical changes [100], and the improvement of sleep quality [125, 126]. However, despite significant advances in this field of research, the risks of this diet and its advantages over alternative strategies are far from being fully elucidated and need further clarification. One of the main gaps in the existing literature is that human studies investigating the mood-stabilizing or anxiolytic effect of the ketogenic diet are largely limited to case studies involved in a small number of individuals. Obviously more research using large-scale human cohorts are needed to

better understand the effect of the ketogenic diet on behavior and biomarkers related to mood and anxiety disorders. Notwithstanding this limitation, the ketogenic diet has shown great efficacy in the management of mood and anxiety disorders and is already extensively being used as a complement or alternative strategy to conventional pharmacotherapy.

## References


1. Masood W, Annamaraju P, Uppaluri KR. Ketogenic diet. In: StatPearls [Internet]. Treasure Island, FL: StatPearls; 2021. https://www.ncbi.nlm.nih.gov/books/NBK499830/. Accessed 22 Nov 2021.
2. Bostock EC, Kirkby KC, Taylor BV. The current status of the ketogenic diet in psychiatry. Front Psych. 2017;8:43. https://doi.org/10.3389/fpsyt.2017.00043.
3. Zhang Y, Xu J, Zhang K, et al. The anticonvulsant effects of ketogenic diet on epileptic seizures and potential mechanisms. Curr Neuropharmacol. 2018;16(1):66–70.
4. Bridge EM, Iob LV. The mechanism of the ketogenic diet in epilepsy. J Ment Sci. 1931;77 (317):437–8.
5. Azar ST. Benefits of ketogenic diet for management of type two diabetes: a review. J Obes Eat Disord. 2016;2:2. https://doi.org/10.21767/2471-8203.100022.
6. Gibson AA, Seimon RV, Lee CM, et al. Do ketogenic diets really suppress appetite? A systematic review and meta-analysis. Obes Rev. 2015;16:64–76. https://doi.org/10.1111/obr.12230.
7. Kong Z, Sun S, Shi Q, et al. Short-term ketogenic diet improves abdominal obesity in overweight/obese Chinese young females. Front Physiol. 2020;11:856. https://doi.org/10.3389/fphys.2020.00856.
8. Samaha FF, Iqbal N, Seshadri P, et al. A low-carbohydrate as compared with a low-fat diet in severe obesity. N Engl J Med. 2003;348(21):2074–81. https://doi.org/10.1056/NEJMoa022637.
9. Aubert G, Martin OJ, Horton JL, et al. The failing heart relies on ketone bodies as a fuel. Circulation. 2016;133(8):698–705. https://doi.org/10.1161/CIRCULATIONAHA.115.017355.
10. De Rosa S, Arcidiacono B, Chiefari E, et al. Type 2 diabetes mellitus and cardiovascular disease: genetic and epigenetic links. Front Endocrinol (Lausanne). 2018;9:2. https://doi.org/10.3389/fendo.2018.00002.
11. Kosinski C, Jornayvaz FR. Effects of ketogenic diets on cardiovascular risk factors: evidence from animal and human studies. Nutrients. 2017;9(5):517. https://doi.org/10.3390/nu9050517.
12. Weber DD, Aminazdeh-Gohari S, Kofler B. Ketogenic diet in cancer therapy. Aging (Albany NY). 2018;10(2):164–5. https://doi.org/10.18632/aging.101382.
13. Tillery E, Ellis K, Threatt T, et al. The use of the ketogenic diet in the treatment of psychiatric disorders. Ment Health Clin. 2021;11:211–9. https://doi.org/10.9740/mhc.2021.05.211.
14. Niepoetter P, Gopalan C. The effects of ketogenic diets on psychiatric disorders involving mitochondrial dysfunction: a literature review of the influence of dieting on autism, depression, anxiety, and schizophrenia. HAPS Educator. 2019;23:426–31. https://doi.org/10.21692/haps.2019.002.
15. Bradford DW, Cunningham N. Psychotic disorders cause the greatest mortality disparity among mental disorders, though more deaths are attributable overall to mood and anxiety disorders. Evid Based Ment Health. 2016;19(2):58. https://doi.org/10.1136/eb-2015-102100.
16. Fakhoury M. New insights into the neurobiological mechanisms of major depressive disorders. Gen Hosp Psychiatry. 2015;37(2):172–7. https://doi.org/10.1016/j.genhosppsych.2015.01.005.

17. Lim GY, Tam WW, Lu Y, et al. Prevalence of depression in the community from 30 countries between 1994 and 2014. Sci Rep. 2018;8(1):2861. https://doi.org/10.1038/s41598-018-21243-x.
18. Fakhoury M. Diagnosis of major depressive disorders: clinical and biological perspectives. In: Kim Y-K, editor. Understanding depression. Singapore: Springer Nature; 2018. https://doi.org/10.1007/978-981-10-6577-4_4.
19. Vieta E, Berk M, Schulze T, et al. Bipolar disorders. Nat Rev Dis Primers. 2018;4:18008. https://doi.org/10.1038/nrdp.2018.8.
20. Jain A, Mitra P. Bipolar affective disorder. In: StatPearls [Internet]. Treasure Island, FL: StatPearls; 2021. https://www.ncbi.nlm.nih.gov/books/NBK558998/.
21. Culpepper L. The diagnosis and treatment of bipolar disorder: decision-making in primary care. Prim Care Companion CNS Disord. 2014;16(3):PCC.13r01609. https://doi.org/10.4088/PCC.13r01609.
22. Weiner M, Warren L, Fiedorowicz JG. Cardiovascular morbidity and mortality in bipolar disorder. Ann Clin Psychiatry. 2011;23(1):40–7.
23. Spoorthy MS, Chakrabarti S, Grover S. Comorbidity of bipolar and anxiety disorders: an overview of trends in research. World J Psychiatry. 2019;9(1):7–29. https://doi.org/10.5498/wjp.v9.i1.7.
24. Fervaha G, Zakzanis KK, Foussias G, et al. Motivational deficits and cognitive test per-formance in schizophrenia. JAMA Psychiat. 2014;71:1058–65.
25. Fakhoury M. Role of the endocannabinoid system in the pathophysiology of schizophrenia. Mol Neurobiol. 2017;54(1):768–78. https://doi.org/10.1007/s12035-016-9697-5.
26. Charlson FJ, Ferrari AJ, Santomauro DF, et al. Global epidemiology and burden of schizophrenia: findings from the global burden of disease study 2016. Schizophr Bull. 2018;44(6):1195–203. https://doi.org/10.1093/schbul/sby058.
27. Lau JY, Eley TC. The genetics of mood disorders. Annu Rev Clin Psychol. 2010;6:313–37. https://doi.org/10.1146/annurev.clinpsy.121208.131308.
28. Sullivan PF, Neale MC, Kendler KS. Genetic epidemiology of major depression: review and meta-analysis. Am J Psychiatry. 2000;157(10):1552–62. https://doi.org/10.1176/appi.ajp.157.10.1552.
29. Witt SH, Streit F, Jungkunz M, et al. Genome-wide association study of borderline personality disorder reveals genetic overlap with bipolar disorder, major depression and schizophrenia. Transl Psychiatry. 2017;7(6):e1155.
30. Leussis MP, Madison JM, Petryshen TL. Ankyrin 3: genetic association with bipolar disorder and relevance to disease pathophysiology. Biol Mood Anxiety Disord. 2012;2:18. https://doi.org/10.1186/2045-5380-2-18.
31. Fiorentino A, O'Brien NL, Locke DP, et al. Analysis of ANK3 and CACNA1C variants identified in bipolar disorder whole genome sequence data. Bipolar Disord. 2014;16(6):583–91. https://doi.org/10.1111/bdi.12203.
32. Moon AL, Haan N, Wilkinson LS, et al. CACNA1C: association with psychiatric disorders, behavior, and neurogenesis. Schizophr Bull. 2018;44(5):958–65. https://doi.org/10.1093/schbul/sby096.
33. Abdolmaleky HM, Cheng KH, Faraone SV, et al. Hypomethylation of MB-COMT promoter is a major risk factor for schizophrenia and bipolar disorder. Hum Mol Genet. 2006;15(21):3132–45. https://doi.org/10.1093/hmg/ddl253.
34. Szczepankiewicz A, Leszczyńska-Rodziewicz A, Pawlak J, et al. Epistatic interaction between CRHR1 and AVPR1b variants as a predictor of major depressive disorder. Psychiatr Genet. 2013;23(6):239–46. https://doi.org/10.1097/YPG.0000000000000007.
35. Fakhoury M. Revisiting the serotonin hypothesis: implications for major depressive disorders. Mol Neurobiol. 2016;53(5):2778–86. https://doi.org/10.1007/s12035-015-9152-z.
36. Ruhé HG, Mason NS, Schene AH. Mood is indirectly related to serotonin, norepinephrine and dopamine levels in humans: a meta-analysis of monoamine depletion studies. Mol Psychiatry. 2007;12(4):331–59. https://doi.org/10.1038/sj.mp.4001949.

37. Brisch R, Saniotis A, Wolf R, et al. The role of dopamine in schizophrenia from a neurobiological and evolutionary perspective: old fashioned, but still in vogue. Front Psych. 2014;5:47. https://doi.org/10.3389/fpsyt.2014.00047.
38. Li P, Snyder GL, Vanover KE. Dopamine targeting drugs for the treatment of schizophrenia: past, present and future. Curr Top Med Chem. 2016;16(29):3385–403. https://doi.org/10.2174/1568026616666160608084834.
39. Niciu MJ, Ionescu DF, Richards EM, et al. Glutamate and its receptors in the pathophysiology and treatment of major depressive disorder. J Neural Transm (Vienna). 2014;121(8):907–24. https://doi.org/10.1007/s00702-013-1130-x.
40. Grossman F, Potter WZ. Catecholamines in depression: a cumulative study of urinary norepinephrine and its major metabolites in unipolar and bipolar depressed patients versus healthy volunteers at the NIMH. Psychiatry Res. 1999;87(1):21–7. https://doi.org/10.1016/s0165-1781(99)00055-4.
41. Ongür D, Drevets WC, Price J. Glial reduction in the subgenual prefrontal cortex in mood disorders. Proc Natl Acad Sci U S A. 1998;95(22):13290–5. https://doi.org/10.1073/pnas.95.22.13290.
42. Bowley MP, Drevets WC, Ongür D, et al. Low glial numbers in the amygdala in major depressive disorder. Biol Psychiatry. 2002;52(5):404–12. https://doi.org/10.1016/s0006-3223(02)01404-x.
43. Schroeter ML, Abdul-Khaliq H, Sacher J, et al. Mood disorders are glial disorders: evidence from in vivo studies. Cardiovasc Psychiatry Neurol. 2010;2010:780645. https://doi.org/10.1155/2010/780645.
44. Pace TW, Hu F, Miller AH. Cytokine-effects on glucocorticoid receptor function: relevance to glucocorticoid resistance and the pathophysiology and treatment of major depression. Brain Behav Immun. 2007;21(1):9–19. https://doi.org/10.1016/j.bbi.2006.08.009.
45. Bonelli RM, Cummings JL. Frontal-subcortical circuitry and behavior. Dialogues Clin Neurosci. 2007;9(2):141–51. https://doi.org/10.31887/DCNS.2007.9.2/rbonelli.
46. Russo SJ, Nestler EJ. The brain reward circuitry in mood disorders. Nat Rev Neurosci. 2013;14(9):609–25. https://doi.org/10.1038/nrn3381.
47. Martin P. The epidemiology of anxiety disorders: a review. Dialogues Clin Neurosci. 2003;5(3):281–98. https://doi.org/10.31887/DCNS.2003.5.3/pmartin.
48. Domschke K, Maron E. Genetic factors in anxiety disorders. Mod Trends Pharmacopsychiatry. 2013;29:24–46. https://doi.org/10.1159/000351932.
49. Platt R, Williams SR, Ginsburg GS. Stressful life events and child anxiety: examining parent and child mediators. Child Psychiatry Hum Dev. 2016;47(1):23–34. https://doi.org/10.1007/s10578-015-0540-4.
50. Goddard AW, Ball SG, Martinez J, et al. Current perspectives of the roles of the central norepinephrine system in anxiety and depression. Depress Anxiety. 2010;27(4):339–50. https://doi.org/10.1002/da.20642.
51. Riaza Bermudo-Soriano C, Perez-Rodriguez MM, Vaquero-Lorenzo C, et al. New perspectives in glutamate and anxiety. Pharmacol Biochem Behav. 2012;100(4):752–74. https://doi.org/10.1016/j.pbb.2011.04.010.
52. Nuss P. Anxiety disorders and GABA neurotransmission: a disturbance of modulation. Neuropsychiatr Dis Treat. 2015;11:165–75. https://doi.org/10.2147/NDT.S58841.
53. Boyle NB, Lawton C, Dye L. The effects of magnesium supplementation on subjective anxiety and stress-a systematic review. Nutrients. 2017;9(5):429. https://doi.org/10.3390/nu9050429.
54. Vogelzangs N, Beekman A, de Jonge P, et al. Anxiety disorders and inflammation in a large adult cohort. Transl Psychiatry. 2013;3:e249. https://doi.org/10.1038/tp.2013.27.
55. Haroon E, Raison CL, Miller AH. Psychoneuroimmunology meets neuropsychopharmacology: translational implications of the impact of inflammation on behavior. Neuropsychopharmacology. 2012;37(1):137–62. https://doi.org/10.1038/npp.2011.205.

56. Michopoulos V, Powers A, Gillespie CF, et al. Inflammation in fear- and anxiety-based disorders: PTSD, GAD, and beyond. Neuropsychopharmacology. 2017;42(1):254–70. https://doi.org/10.1038/npp.2016.146.
57. Felger JC. Imaging the role of inflammation in mood and anxiety-related disorders. Curr Neuropharmacol. 2018;16(5):533–58. https://doi.org/10.2174/1570159X15666171123201142.
58. Baker DG, Ekhator NN, Kasckow JW, et al. Plasma and cerebrospinal fluid interleukin-6 concentrations in posttraumatic stress disorder. Neuroimmunomodulation. 2001;9(4):209–17. https://doi.org/10.1159/000049028.
59. Kim TD, Lee S, Yoon S. Inflammation in post-traumatic stress disorder (PTSD): a review of potential correlates of PTSD with a neurological perspective. Antioxidants (Basel). 2020;9(2):107. https://doi.org/10.3390/antiox9020107.
60. Pace TW, Wingenfeld K, Schmidt I, et al. Increased peripheral NF-κB pathway activity in women with childhood abuse-related posttraumatic stress disorder. Brain Behav Immun. 2012;26(1):13–7. https://doi.org/10.1016/j.bbi.2011.07.232.
61. Smith AK, Conneely KN, Kilaru V, et al. Differential immune system DNA methylation and cytokine regulation in post-traumatic stress disorder. Am J Med Genet B Neuropsychiatr Genet. 2011;156B(6):700–8. https://doi.org/10.1002/ajmg.b.31212.
62. Rusiecki JA, Byrne C, Galdzicki Z, et al. PTSD and DNA methylation in select immune function gene promoter regions: a repeated measures case-control study of U.S. military service members. Front Psych. 2013;4:56. https://doi.org/10.3389/fpsyt.2013.00056.
63. Vgontzas AN, Papanicolaou DA, Bixler EO, et al. Circadian interleukin-6 secretion and quantity and depth of sleep. J Clin Endocrinol Metab. 1999;84(8):2603–7. https://doi.org/10.1210/jcem.84.8.5894.
64. Meier-Ewert HK, Ridker PM, Rifai N, et al. Effect of sleep loss on C-reactive protein, an inflammatory marker of cardiovascular risk. J Am Coll Cardiol. 2004;43(4):678–83. https://doi.org/10.1016/j.jacc.2003.07.050.
65. Muscatell KA, Dedovic K, Slavich GM, et al. Greater amygdala activity and dorsomedial prefrontal-amygdala coupling are associated with enhanced inflammatory responses to stress. Brain Behav Immun. 2015;43:46–53. https://doi.org/10.1016/j.bbi.2014.06.201.
66. Wignall EL, Dickson JM, Vaughan P, et al. Smaller hippocampal volume in patients with recent-onset posttraumatic stress disorder. Biol Psychiatry. 2004;56(11):832–6. https://doi.org/10.1016/j.biopsych.2004.09.015.
67. Woon FL, Sood S, Hedges DW. Hippocampal volume deficits associated with exposure to psychological trauma and posttraumatic stress disorder in adults: a meta-analysis. Prog Neuropsychopharmacol Biol Psychiatry. 2010;34(7):1181–8. https://doi.org/10.1016/j.pnpbp.2010.06.016.
68. Milham MP, Nugent AC, Drevets WC, et al. Selective reduction in amygdala volume in pediatric anxiety disorders: a voxel-based morphometry investigation. Biol Psychiatry. 2005;57(9):961–6. https://doi.org/10.1016/j.biopsych.2005.01.038.
69. Rogers MA, Yamasue H, Abe O, et al. Smaller amygdala volume and reduced anterior cingulate gray matter density associated with history of post-traumatic stress disorder. Psychiatry Res. 2009;174(3):210–6. https://doi.org/10.1016/j.pscychresns.2009.06.001.
70. Fonzo GA, Simmons AN, Thorp SR, et al. Exaggerated and disconnected insular-amygdalar blood oxygenation level-dependent response to threat-related emotional faces in women with intimate-partner violence posttraumatic stress disorder. Biol Psychiatry. 2010;68(5):433–41. https://doi.org/10.1016/j.biopsych.2010.04.028.
71. Taggart AK, Kero J, Gan X, et al. (D)-beta-Hydroxybutyrate inhibits adipocyte lipolysis via the nicotinic acid receptor PUMA-G. J Biol Chem. 2005;280(29):26649–52. https://doi.org/10.1074/jbc.C500213200.
72. Yang H, Shan W, Zhu F, et al. Ketone bodies in neurological diseases: focus on neuroprotection and underlying mechanisms. Front Neurol. 2019;10:585. https://doi.org/10.3389/fneur.2019.00585.

73. Rahman M, Muhammad S, Khan MA, et al. The β-hydroxybutyrate receptor HCA2 activates a neuroprotective subset of macrophages. Nat Commun. 2014;5:3944. https://doi.org/10.1038/ncomms4944.
74. Rusek M, Pluta R, Ułamek-Kozioł M, et al. Ketogenic diet in Alzheimer's disease. Int J Mol Sci. 2019;20(16):3892. https://doi.org/10.3390/ijms20163892.
75. Cullingford TE. The ketogenic diet; fatty acids, fatty acid-activated receptors and neurological disorders. Prostaglandins Leukot Essent Fatty Acids. 2004;70(3):253–64. https://doi.org/10.1016/j.plefa.2003.09.008.
76. Yang X, Cheng B. Neuroprotective and anti-inflammatory activities of ketogenic diet on MPTP-induced neurotoxicity. J Mol Neurosci. 2010;42(2):145–53. https://doi.org/10.1007/s12031-010-9336-y.
77. Dupuis N, Curatolo N, Benoist JF, et al. Ketogenic diet exhibits anti-inflammatory properties. Epilepsia. 2015;56(7):e95–8. https://doi.org/10.1111/epi.13038.
78. Youm YH, Nguyen KY, Grant RW, et al. The ketone metabolite β-hydroxybutyrate blocks NLRP3 inflammasome-mediated inflammatory disease. Nat Med. 2015;21(3):263–9. https://doi.org/10.1038/nm.3804.
79. Jeong EA, Jeon BT, Shin HJ, et al. Ketogenic diet-induced peroxisome proliferator-activated receptor-γ activation decreases neuroinflammation in the mouse hippocampus after kainic acid-induced seizures. Exp Neurol. 2011;232(2):195–202. https://doi.org/10.1016/j.expneurol.2011.09.001.
80. Forsythe CE, Phinney SD, Fernandez ML, et al. Comparison of low fat and low carbohydrate diets on circulating fatty acid composition and markers of inflammation. Lipids. 2008;43(1):65–77. https://doi.org/10.1007/s11745-007-3132-7.
81. Koh S, Dupuis N, Auvin S. Ketogenic diet and Neuroinflammation. Epilepsy Res. 2020;167:106454. https://doi.org/10.1016/j.eplepsyres.2020.106454.
82. Kim DY, Hao J, Liu R, et al. Inflammation-mediated memory dysfunction and effects of a ketogenic diet in a murine model of multiple sclerosis. PLoS One. 2012;7(5):e35476. https://doi.org/10.1371/journal.pone.0035476.
83. Ruskin DN, Sturdevant IC, Wyss LS, et al. Ketogenic diet effects on inflammatory allodynia and ongoing pain in rodents. Sci Rep. 2021;11(1):725. https://doi.org/10.1038/s41598-020-80727-x.
84. Salim S, Chugh G, Asghar M. Inflammation in anxiety. Adv Protein Chem Struct Biol. 2012;88:1–25. https://doi.org/10.1016/B978-0-12-398314-5.00001-5.
85. Enache D, Pariante CM, Mondelli V. Markers of central inflammation in major depressive disorder: a systematic review and meta-analysis of studies examining cerebrospinal fluid, positron emission tomography and post-mortem brain tissue. Brain Behav Immun. 2019;81:24–40. https://doi.org/10.1016/j.bbi.2019.06.015.
86. Fries GR, Walss-Bass C, Bauer ME, et al. Revisiting inflammation in bipolar disorder. Pharmacol Biochem Behav. 2019;177:12–9. https://doi.org/10.1016/j.pbb.2018.12.006.
87. Müller N. Inflammation in schizophrenia: pathogenetic aspects and therapeutic considerations. Schizophr Bull. 2018;44(5):973–82. https://doi.org/10.1093/schbul/sby024.
88. Zhao Y, Ma R, Shen J, et al. A mouse model of depression induced by repeated corticosterone injections. Eur J Pharmacol. 2008;581:113–20. https://doi.org/10.1016/j.ejphar.2007.12.005.
89. Golden S, Covington H, Berton O, et al. A standardized protocol for repeated social defeat stress in mice. Nat Protoc. 2011;6:1183–91. https://doi.org/10.1038/nprot.2011.361.
90. Yoshida K, Drew M, Kono A, et al. Chronic social defeat stress impairs goal-directed behavior through dysregulation of ventral hippocampal activity in male mice. Neuropsychopharmacology. 2021;46:1606–16. https://doi.org/10.1038/s41386-021-00990-y.
91. Sarnyai Z, Kraeuter AK, Palmer CM. Ketogenic diet for schizophrenia: clinical implication. Curr Opin Psychiatry. 2019;32(5):394–401. https://doi.org/10.1097/YCO.0000000000000535.

92. Włodarczyk A, Wiglusz M, Cubała W. Ketogenic diet for schizophrenia: nutritional approach to antipsychotic treatment. Med Hypotheses. 2018;118:74–7. https://doi.org/10.1016/j.mehy.2018.06.022.
93. Sullivan C, Mielnik C, Funk A, et al. Measurement of lactate levels in postmortem brain, iPSCs, and animal models of schizophrenia. Sci Rep. 2019;9:5087. https://doi.org/10.1038/s41598-019-41572-9.
94. Sullivan CR, O'Donovan SM, McCullumsmith RE, et al. Defects in bioenergetic coupling in schizophrenia. Biol Psychiatry. 2018;83(9):739–50. https://doi.org/10.1016/j.biopsych.2017.10.014.
95. Yuksel C, Chen X, Chouinard VA, et al. Abnormal brain bioenergetics in first-episode psychosis. Schizophr Bull Open. 2021;2(1):sgaa073. https://doi.org/10.1093/schizbullopen/sgaa073.
96. Brietzke E, Mansur R, Subramaniapillai M, et al. Ketogenic diet as a metabolic therapy for mood disorders: evidence and developments. Neurosci Biobehav Rev. 2018;94:11–6. https://doi.org/10.1016/j.neubiorev.2018.07.020.
97. Murphy P, Likhodii S, Nylen K, et al. The antidepressant properties of the ketogenic diet. Biol Psychiatry. 2004;56:981–3. https://doi.org/10.1016/j.biopsych.2004.09.019.
98. Stafstrom C, Rho J. The ketogenic diet as a treatment paradigm for diverse neurological disorders. Front Pharmacol. 2012;3:59. https://doi.org/10.3389/fphar.2012.00059.
99. Guan YF, Huang GB, Xu MD. Anti-depression effects of ketogenic diet are mediated via the restoration of microglial activation and neuronal excitability in the lateral habenula. Brain Behav Immun. 2020;88:748–62. https://doi.org/10.1016/j.bbi.2020.05.032.
100. Sussman D, Germann J, Henkelman M. Gestational ketogenic diet programs brain structure and susceptibility to depression & anxiety in the adult mouse offspring. Brain Behav. 2015;5(2):e00300. https://doi.org/10.1002/brb3.300.
101. Gould T, Einat H. Animal models of bipolar disorder and mood stabilizer efficacy: a critical need for improvement. Neurosci Biobehav Rev. 2007;31:825–31. https://doi.org/10.1016/j.neubiorev.2007.05.007.
102. Grunze H. Bipolar disorder. In: Neurobiology of brain disorders; 2015. p. 655–73. https://doi.org/10.1016/b978-0-12-398270-4.00040-9.
103. Phelps J, Siemers S, El-Mallakh R. The ketogenic diet for type II bipolar disorder. Neurocase. 2013;19:423–6. https://doi.org/10.1080/13554794.2012.690421.
104. Dsouza A, Haque S, Aggarwal R. The influence of ketogenic diets on mood stability in bipolar disorder. Asian J Psychiatr. 2017;41:86–7. https://doi.org/10.1016/j.ajp.2017.10.024.
105. Operto FF, Matricardi S, Pastorino GMG, et al. The ketogenic diet for the treatment of mood disorders in comorbidity with epilepsy in children and adolescents. Front Pharmacol. 2020;11:578396. https://doi.org/10.3389/fphar.2020.578396. PMID: 33381032; PMCID: PMC7768824
106. Campbell I, Campbell H. Ketosis and bipolar disorder: controlled analytic study of online reports. BJPsych Open. 2019;5(4):e58. https://doi.org/10.1192/bjo.2019.49.
107. Yaroslavsky Y, Stahl Z, Belmaker R. Ketogenic diet in bipolar illness. Bipolar Disord. 2002;4:75. https://doi.org/10.1034/j.1399-5618.2002.01212.x.
108. El-Mallakh RS, Paskitti ME. The ketogenic diet may have mood-stabilizing properties. Med Hypotheses. 2001;57(6):724–6. https://doi.org/10.1054/mehy.2001.1446.
109. Palmer C, Gilbert-Jaramillo J, Westman E. The ketogenic diet and remission of psychotic symptoms in schizophrenia: two case studies. Schizophr Res. 2019;208:439–40. https://doi.org/10.1016/j.schres.2019.03.019.
110. Palmer CM. Ketogenic diet in the treatment of schizoaffective disorder: two case studies. Schizophr Res. 2017;189:208–9. https://doi.org/10.1016/j.schres.2017.01.053.
111. Kraft BD, Westman EC. Schizophrenia, gluten, and low-carbohydrate, ketogenic diets: a case report and review of the literature. Nutr Metab (Lond). 2009;6:10. https://doi.org/10.1186/1743-7075-6-10.

112. Cox N, Gibas S, Salisbury M, et al. Ketogenic diets potentially reverse type II diabetes and ameliorate clinical depression: a case study. Diabetes Metab Syndr. 2019;13(2):1475–9. https://doi.org/10.1016/j.dsx.2019.01.055.
113. Ströhle A, Gensichen J, Domschke K. The diagnosis and treatment of anxiety disorders. Dtsch Arztebl Int. 2018;155(37):611–20. https://doi.org/10.3238/arztebl.2018.0611.
114. Mahan PL, Mahan MP, Park NJ, et al. Work environment stressors, social support, anxiety, and depression among secondary school teachers. AAOHN J. 2010;58(5):197–205. https://doi.org/10.3928/08910162-20100416-01.
115. Shensa A, Sidani JE, Dew MA, et al. Social media use and depression and anxiety symptoms: a cluster analysis. Am J Health Behav. 2018;42(2):116–28. https://doi.org/10.5993/AJHB.42.2.11.
116. Cuijpers P, Sijbrandij M, Koole SL, et al. The efficacy of psychotherapy and pharmacotherapy in treating depressive and anxiety disorders: a meta-analysis of direct comparisons. World Psychiatry. 2013;12(2):137–48. https://doi.org/10.1002/wps.20038.
117. Norwitz NG, Naidoo U. Nutrition as metabolic treatment for anxiety. Front Psych. 2021;12:598119. https://doi.org/10.3389/fpsyt.2021.598119.
118. Włodarczyk A, Cubała WJ, Wielewicka A. Ketogenic diet: a dietary modification as an anxiolytic approach? Nutrients. 2020;12(12):3822. https://doi.org/10.3390/nu12123822.
119. Lydiard RB. The role of GABA in anxiety disorders. J Clin Psychiatry. 2003;64(Suppl 3):21–7.
120. Calderón N, Betancourt L, Hernández L, et al. A ketogenic diet modifies glutamate, gamma-aminobutyric acid and agmatine levels in the hippocampus of rats: a microdialysis study. Neurosci Lett. 2017;642:158–62. https://doi.org/10.1016/j.neulet.2017.02.014.
121. Mokler DJ, Torres OI, Galler JR, et al. Stress-induced changes in extracellular dopamine and serotonin in the medial prefrontal cortex and dorsal hippocampus of prenatally malnourished rats. Brain Res. 2007;1148:226–33. https://doi.org/10.1016/j.brainres.2007.02.031.
122. Alamy M, Bengelloun WA. Malnutrition and brain development: an analysis of the effects of inadequate diet during different stages of life in rat. Neurosci Biobehav Rev. 2012;36(6):1463–80. https://doi.org/10.1016/j.neubiorev.2012.03.009.
123. Ari C, Kovács Z, Juhasz G, et al. Exogenous ketone supplements reduce anxiety-related behavior in Sprague-Dawley and Wistar albino Glaxo/Rijswijk rats. Front Mol Neurosci. 2016;9:137. https://doi.org/10.3389/fnmol.2016.00137.
124. Oh CM, Kim HY, Na HK, et al. The effect of anxiety and depression on sleep quality of individuals with high risk for insomnia: a population-based study. Front Neurol. 2019;10:849. https://doi.org/10.3389/fneur.2019.00849.
125. Hallböök T, Lundgren J, Rosén I. Ketogenic diet improves sleep quality in children with therapy-resistant epilepsy. Epilepsia. 2007;48(1):59–65. https://doi.org/10.1111/j.1528-1167.2006.00834.x.
126. Ünalp A, Baysal BT, Sarıtaş S, et al. Evaluation of the effects of ketogenic diet therapy on sleep quality in children with drug-resistant epilepsy and their mothers. Epilepsy Behav. 2021;124:108327. https://doi.org/10.1016/j.yebeh.2021.108327.