51. Alzheimer's Disease

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Alzheimer' s disease (AD) is the most common neurodegenerative disorder in late life which is clinically characterized by dementia and progressive cognitive impairments with presently no effective treatment. This chapter summarizes recent progress achieved during the last decades in understanding the pathogenesis of AD. Basing on the pathomorphological hallmarks (senile amyloid plaque deposits, occurance of neurofibrillary tangles as hyperphosphorylated tau protein in cerebral cortex and hippocampus) and other consistent features of the disease (neurodegeneration, cholinergic dysfunction, vascular impairments), the mayor hypotheses of cause and development of the sporadic, not genetically inherited, AD are described. Finally, to reflect the disease in its entirety and internal connective relationships, the different pathogenetic hypotheses are tentatively combined to describe the interplay of the essential features of AD and their mutually influencing pathogenetic processes in a unified model. Such a unified approach may provide a basis to model pathogenesis and progression of AD by application of computational methods such as the recently introduced novel research framework for building probabilistic computational neurogenetic models (pCNGM) by *Kasabov* and coworkers [51.1].

51.1 Alzheimer's Disease

51.1.1 Epidemiology

In 1906/1907 the German psychiatrist Alois Alzheimer described a patient with progressive memory impairment, disordered cognitive function, altered personality and social behavior, including paranoia, delusions, and loss of social appropriateness. Following autopsy, in the patient's cerebral cortex and hippocampus he observed abundant extracellular amyloid-like protein deposits (neuritic plaques) and a number of pyramidal neurons containing intracellular inclusions of fibrillary structures (neurofibrillary tangles), which was accompanied by considerable neuronal, in particular cholinergic, cell loss [51.2]. During the following years similar cases were observed, and already in the German textbook of psychiatry edited by Emil Kraepelin in 1910, this novel brain disorder was named after *Alois Alzheimer*. Later it became evident that this kind of disorder also provides a common basis for impairments in cognition in elderly people in late life. The modern era of AD research, however, began with the identification of the β-amyloid sequence in the congophilic angiopathy of AD and Down's syndrome [51.3]. The finding that the amyloid plaques observed in both disorders contained largely the same peptide [51.4] and the cloning of the β-amyloid precursor protein gene [51.5–8] have led to major progress in understanding the pathology and biochemistry of AD.

AD represents the most common neurodegenerative disease with dementia and progressing cognitive deficits. Presently, in Germany there are about 1.2 million people suffering from AD, while worldwide there are estimates of more than 35 million AD patients [51.9]. Aging is the most important risk factor for AD (for a review, see [51.10]). At the age of 65 years about 1.2% of the population is afflicted, with an approximate doubling of incidence for every 5 years of age afterward. For those over the age of 90, the prevalence increases to nearly 40%. Due to ever-increasing life expectancy, the number of affected individuals is projected to rise to more than 2.3 million in Germany [51.9] and about 100 million worldwide by 2050 [51.11]. The increasing number of elderly people suffering from cognitive impairment and dementia will produce a considerable economic burden for the social communities in the near future, due to the growing costs of healthcare and welfare systems. This challenging perspective indicates that detailed knowledge of the pathogenetic mechanisms of AD as well as pharmacotherapeutical strategies to combat AD are urgently required. It has been estimated that a delay in the onset of AD by 5 years may decrease the number of affected people by half.

51.1.2 Morphopathological Hallmarks

The pathology of AD is characterized by two major histopathological hallmarks such as senile plaques (syn. neuritic plaques, amyloid plaques) deposed extracellularly in cerebral cortical and hippocampal areas, and neurofibrillary tangles that occupy much of the cytoplasm of select cortical pyramidal neurons. These neuropathological hallmarks of AD are particularly prominent in areas such as the parietal and temporal cortices, the hippocampus, the entorhinal cortex, and the amygdala. In addition to senile plaques and neurofibrillary tangles, brains from AD patients also demonstrate astrocytic gliosis, reactive microglia, inflammation, as well as neuronal cell loss and synaptic dysfunction, which have been assumed to be consequences of the accumulation of the pathological protein components (for a review, see [51.12]).

Senile plaques are extracellular deposits that are mostly composed of β-amyloid (Aβ), a proteolytic fragment of the APP. The deposits are associated with neuronal terminals and degenerative swollen neurites and are surrounded by a web of astrocytic processes and microglia cells [51.3, 4, 13].

Neurofibrillary tangles, the second histopathological feature in AD, are pathological protein aggregates detectable in the cytoplasm of select cortical pyramidal neurons, which mostly consist of hyperphosphorylated tau, a microtubule-associated protein ([51.14–16], see also [51.17]). The tau protein is a highly soluble protein that functions as a modulator of the stability and flexibility of axonal microtubules, mediated by its degree of phosphorylation. Hyperphosphorylation of tau depresses its microtubule assembly activity and its binding to microtubules [51.18]. Abnormal hyperphosphorylated tau demonstrates a high tendency to aggregate and to form paired helical filaments (PHF) and straight filaments (SF), thus causing insoluble cytoplasmic inclusions, which disrupt the structure and function of

the neurons [51.19]. However, recent studies have provided evidence that prefibrillar, oligomeric forms of tau, rather than PHF, cause the neurodegeneration observed in AD [51.20].

Many phosphokinases including glycogen synthase kinase 3β (GSK3β), cyclin-dependent kinase 5 (cdk5), and extracellular signal-related kinase 2 (ERK2), as well as the dual specificity tyrosine-phosphorylation-regulated kinase1A (DYRK1A) have been assumed to be involved in tau phosphorylation (for a review, see [51.21]).

The level of hyperphosphorylated tau in AD brains is about four–eight times higher as compared to age-matched normal brains. In the adult human brain, six tau isoforms are expressed through alternative splicing of a single tau gene on chromosome 17 (for a review, see [51.22]). Three isoforms contain three tandem microtubule binding repeats, while the other three forms contain four microtubule binding repeats. In AD all of the six tau isoforms contribute to the formation of PHF by hyperphosphorylation [51.23].

Tau pathology is also seen in a number of other human neurodegenerative disorders associated with neurodegeneration and dementia [51.22].

There is experimental evidence of a link between Aβ and tau pathology in AD with Aβ deposition preceding the tau pathology [51.21]. Several hypotheses have been suggested, including the *dual pathway* model of causality, whereby Aβ and tau are linked by separate mechanisms driven by a common upstream driver [51.24].

51.1.3 Genetic Factors Predisposing AD

A small subset of AD cases ($< 5\%$) results from an inherited autosomal dominant gene mutations and have an early-onset at ages between 40 and 60 years, while the majority of AD cases are sporadic with a complex aetiology due to interactions between environmental conditions and specific genetic features of the individual [51.23].

In patients with the early-onset familial form of the disease (FAD) that tends to be more aggressive, mutations in genes located on chromosome 21, 14, and 1, encoding the amyloid precursor protein (APP), presenilin1, and presenilin2, respectively, have been found (for reviews, see [51.12,25]), while hitherto no tau gene mutations were observed in AD [51.23].

Moreover, a number of potential risk genes for AD have been described (for a review, see [51.21]), with ApoE being the most consistently associated risk gene. Carriers of the ApoE ε4-allele have a several times higher risk of developing AD as compared with those carrying the ApoE ε4-allele [51.26–28].

Inherited variants in the neuronal sortilin-related sorting receptor SORL1 have been found to be associated with late-onset AD [51.29]. Recently performed genome-wide association studies in late-onset AD patients revealed the identification of nine further novel genes that are linked to immune system function, cholesterol metabolism, and synaptic cell membrane processes, and which explain around 50% of late-onset AD genetics (for a review, see [51.30]).

51.2 Cholinergic Dysfunction in Alzheimer's Disease

Another consistent feature of AD is a progressive neuronal cell loss that is associated with region-specific brain atrophy. In particular, the cholinergic projection from the nucleus basalis of Meynert to areas of the cerebral cortex is the pathway that is very early and most severely affected in brains from AD patients. Changes in the cholinergic transmission in AD have been documented by assessing the major processes occurring at a cholinergic synapse. In the presynaptic compartment of the cholinergic neuron the neurotransmitter acetylcholine is synthesized from choline and acetyl-CoA by the catalytic action of the choline acetyltransferase (ChAT), and subsequently taken up into the vesicle by means of the vesicular acetylcholine transporter (VAChT). Following the action potential-induced release into the synaptic cleft, acetylcholine binds to both post and presynaptically localized cholinergic muscarinic (mAChR) and nicotinic acetylcholine receptors (nAChR). While M1/M3-mAChR subtypes are mainly localized postsynaptically, parts of the M2/M4-mAChR subtypes act as autoreceptors on cholinergic presynaptic compartments to control acetylcholine release. Similarly, nAChRs are predominantly localized on both cholinergic and non-cholinergic nerve terminals to regulate transmitter release. In the synaptic cleft, acetylcholine undergoes a fast degradation by the catalytic action of the acetylcholinesterase (AChE, see also Fig. 51.1).

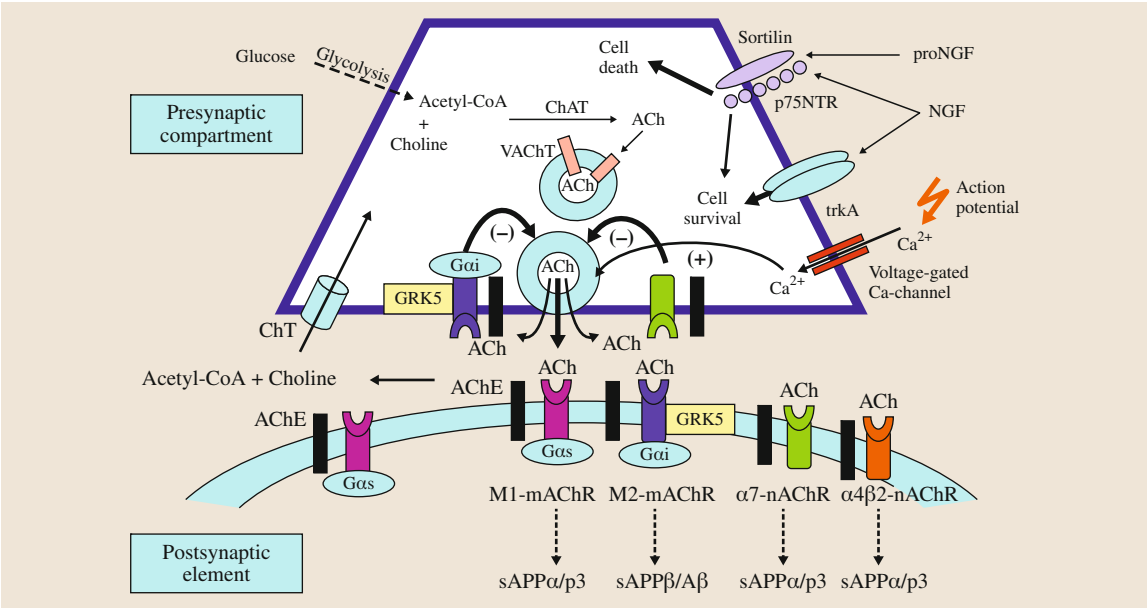

Fig. 51.1 *Depiction of main processes occurring at a cholinergic synapse in the brain.* In the presynaptic compartment of the cholinergic neuron the neurotransmitter acetylcholine is synthesized from choline and acetyl-CoA by the catalytic action of the choline acetyltransferase (ChAT) and subsequently taken up into the vesicle by means of the vesicular acetylcholine transporter (VAChT). Following the action potential-induced, Ca^{2+}-dependent release into the synaptic cleft, acetylcholine (ACh) binds to both post and presynaptically localized cholinergic muscarinic (mAChR) and nicotinic acetylcholine receptors (nAChR). While M1-mAChR subtypes are mainly localized postsynaptically, parts of the M2-mAChR subtypes act as autoreceptors on cholinergic presynaptic compartments to control ACh release. Similarly, nAChRs are predominantly localized on both cholinergic and non-cholinergic nerve terminals to regulate transmitter release. In the synaptic cleft, ACh undergoes a fast degradation by the catalytic action of the acetylcholinesterase (AChE). While activation of M1-mAChR and nAChR favor the non-amyloidogenic route of amyloid precursor protein (APP) processing, stimulation of M2-mAChR drives the APP processing toward the amyloidogenic path. Activated M2-mAChR are selectively desensitized by phosphorylation under the catalytic action of G protein-coupled receptor kinase 5 (GRK5). β-Amyloid (Aβ)-induced GRK5 deficiency leads to presynaptic M2-mAChR hyperactivity, which initiates a cascade of pathological events including enhanced Aβ formation finally resulting in cholinergic dysfunction. For survival and maintenance, cholinergic neurons require neurotrophic support by the nerve growth factor (NGF) mediated through binding to both high- (trkA) and low-affinity (p75NTR) receptors which are nearly exclusively located on central cholinergic cells

For survival and maintenance, cholinergic neurons require neurotrophic support by the nerve growth factor (NGF) mediated through binding to both high- (trkA) and low-affinity (p75NTR) receptors, which are nearly exclusively located on central cholinergic cells (Sect. 51.4). Therefore, NGF and its receptors have additionally been used as markers to further characterize cholinergic dysfunction in AD.

Severe deficits of presynaptic cholinergic markers in the cerebral cortex of patients with early-onset AD were already observed in the late 1970s in a number of studies [51.31–34]. Biochemical and in situ hybridization studies reported a marked and region-dependent loss in ChAT activity (from 30 to 90%) and ChAT mRNA levels (about 50%) in the temporal lobe and frontal and parietal cortices of the AD brain being consistent with striking reductions in the number of basal forebrain cholinergic neurons. Accompanying the degenerations of cholinergic neurons are the presence of neuritic plaques in cholinoceptive cortical target regions, as well as within the basal forebrain nuclei (for references, see e.g., [51.35]). Neurochemical analysis have further revealed deficits in several other presynaptic cholinergic markers, such as acetylcholine synthesis and release, as well as a decreased number of nAChRs. While most studies did not detect changes in M1-

mAChRs, a decrease in the number of M2-mAChRs was reported [51.36].

The correlation of clinical dementia ratings with the reductions in a number of cortical cholinergic markers such as ChAT, M2-mAChR, and nAChR binding, as well as levels of acetylcholine [51.36–38], suggested an association of cholinergic hypofunction with cognitive deficits, which led to the formulation of the cholinergic hypothesis of memory dysfunction in senescence and in AD [51.39].

While there is no doubt that severe loss of cortical cholinergic innervation exists already in the initial stages of presenile (early-onset) as well as in the advanced stages of late-onset AD, the assumption of cholinergic denervation being an early and initial stage also in mild, late-onset AD has been debated (see e.g., [51.40]). Only mild losses of AChE activities have been observed in patients with mild cognitive impairment (MCI, a prodromal stage of AD), and early forms of AD, while in a number of brain regions studied in MCI patients no decrease in ChAT activity has been observed. Similarly, the number of ChAT-positive and VAChT-positive cells was unaltered in MCI as compared to non-demented controls. While the number of ChAT-positive neurons was unchanged, the trkA and p75NTR-containing neurons, which co-localize with ChAT, were significantly reduced in the nucleus basalis of subjects with MCI as compared to those with no cognitive deteriorations. These findings suggest a downregulation of trkA and p75NTR receptors, thus a dysfunction of cholinergic neurons rather than cholinergic cell loss [51.41]. This is further emphasized by observations that other parameters of cholinergic function such as acetylcholine release, high-affinity choline uptake, and expression of mAChR and nAChR are also altered in MCI and early AD [51.35].

Gene expression analysis of single basal forebrain cholinergic neurons revealed that trkA, but not p75NTR, is reduced in MCI. The NGF precursor, proNGF, has been observed to be increased in the cortex of MCI and AD. As proNGF accumulates in the presence of reduced cortical trkA and sustained levels of p75NTR, a shift in the balance between cell survival and death molecules may occur in early AD. Similarly, BDNF and proBDNF, are reduced in the cortex of MCI, further depriving basal forebrain cholinergic cells of trophic support [51.41]. ProNGF is released from the cerebral cortex in an activity-dependent manner together with enzymes required to generate mature NGF. Thus, the upregulation of proNGF observed in AD may indicate a dysregulation in the maturation of NGF leading to enhanced vulnerability of the cholinergic system in AD [51.42]. These data further support the suggestion of a key role of the cholinergic system in the functional processes that lead to AD, while provoking the question whether the gradual progressive loss of cholinergic function is associated with Aβ and tau pathology, as will be discussed in Sect. 51.3.2.

51.3 APP Metabolism and Its Regulation by Cholinergic Mechanisms

51.3.1 Processing of APP and Generation of Aβ

The major constituent of neuritic plaques is the 4 kDa Aβ peptide that is derived from a much larger protein, the APP. APP belongs to a family of glycosylated type-I transmembrane proteins which are ubiquitously expressed but most abundantly in the brain. There are three major isoforms APP695, APP751, and APP770 (containing 696, 751, 770 amino acids, respectively) arising from alternative splicing. APP770 (and APP751) consists of a large extracellular domain, containing a 56 amino acid Kunitz protease inhibitor (KPI) region and glycosylation sites, a single membrane-spanning region, and a shorter intracellular carboxyl terminus. While APP751 and APP770 are expressed in most tissues, APP695 is predominantly expressed in neurons and lacks the KPI domain (for a review, see [51.43]).

The precursor protein contains the 39–42 amino acid sequence of Aβ and is processed either by an α-secretory or β-secretory pathway yielding non-amyloidogenic cleavage products or potentially amyloidogenic Aβ peptides, respectively. In order to generate Aβ the APP must be cleaved by two proteases that have been termed β-and γ-secretases. The β-secretase cleavage results in a truncated APP that is denoted as secretory sAPP β, and in a remaining part consisting of 99 amino acids, the β-carboxy-terminal fragment, β CTF (12 kDa). The β CTF may undergo a further cleavage by the γ-secretase at the amino acid 711 or 713 to release either Aβ (1–40), the predominant species, or to a lesser extent, the more amyloidogenic Aβ (1–42) peptide, respectively, as well as the remaining intracellular domain of APP (AICD). Recently, further APP cleavage sites of the γ-secretase have been described: the ζ-site generating Aβ (1–46) and

the ε-site producing Aβ (1–49). An overview of the amyloidogenic route of APP processing is depicted in Fig. 51.2a.

The released AICD has been shown to regulate transcription of a number of genes including APP, glycogen synthase kinase (GSK) 3β, neprilysin, BACE1, p53, epidermal growth factor receptor (EGFR), and low density lipoprotein receptor-related protein 1 (LRP1), while AICD as the intracellular domain mediates interaction of APP with various cytosolic factors.

Recently, sAPPβ has been reported to undergo a further cleavage by a still unknown mechanism to release N-APP. N-APP was found to bind to the death receptor DR6 to cause axon pruning and neuronal cell death via caspase-6 and -3, respectively [51.44]. A novel function of sAPPβ as a regulator of transthyretin and Klotho gene expression has been detected recently [51.45].

The non-amyloidogenic secretory pathway includes the proteolytic cleavage of APP by the α-secretase between amino acid position 16 and 17 of the Aβ domain, which results in the secretion of a soluble ectodomain of APP, called sAPPα. The remaining membrane-anchored C-terminal fragment of APP, α CTF (83 amino acid residues) may further undergo a γ-secretase cleavage to produce a peptide called p3 and the remaining AICD (see also Fig. 51.2b).

sAPPα has been shown to be involved in mediating neuronal plasticity and survival, protection against excitoxicity, neural stem cell proliferation, and inhibition of stress-induced cdk5 activation; the physiological function of the rapidly degraded p3 peptide is still unclear [51.43].

The α, β and γ-secretases have recently been cloned and identified. The α-secretase is a zinc metalloproteinase and acts as a membrane-bound endoprotease to cleave APP within the plasma membrane. Three members of the ADAM (a disintegrin and metalloproteinase) family have been proposed to act as α-secretase: ADAM9, 10, and 17 (for reviews, see [51.43, 46].

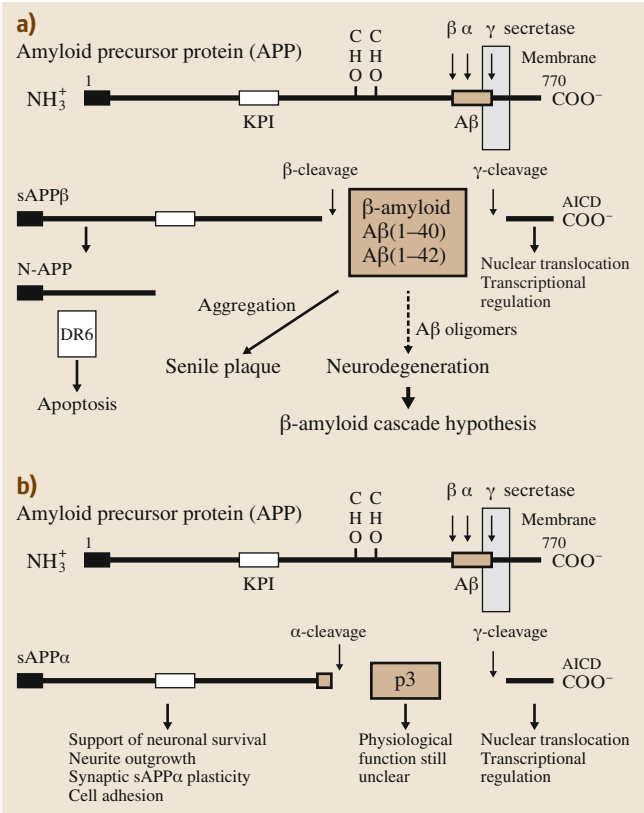

Fig. 51.2a,b *Processing of the amyloid precursor protein (APP).* **(a)** The APP contains the 40–42 amino acid sequence of Aβ, which is partly localized within the transmembrane domain of APP. To generate Aβ the APP is cleaved by two proteases termed as β-and γ-secretases. The β-secretase cleavage results in formation of secretory sAPPβ, and in a remaining part, the β-carboxy-terminal fragment, βCTF. The βCTF may undergo a further cleavage by the γ-secretase to release either Aβ (1–40), the predominant species, or to a lesser extent, the more amyloidogenic Aβ (1–42) peptide, respectively, as well as the remaining intracellular domain of APP (AICD). Aβ peptides may accumulate in the brain as synaptotoxic and cytotoxic oligomers and as fibrillar aggregates in the form of senile plaques. The released AICD has been shown to regulate transcription of a number of genes, while AICD as intracellular domain mediates interaction of APP with various cytosolic factors. Recently, sAPPβ was reported to undergo a further cleavage to release N-APP, which binds to the death receptor DR6 thereby causing axon pruning and neuronal cell death. **(b)** The non-amyloidogenic secretory pathway includes the proteolytic cleavage of APP by the α-secretase between amino acid position 16 and 17 of the Aβ domain which results in the secretion of the soluble ectodomain, sAPPα. The remaining membrane-anchored C-terminal fragment, αCTF may further undergo a γ-secretase cleavage to produce a peptide called p3 and the remaining AICD. sAPPα has been shown to be involved in mediating neuronal plasticity and survival, protection against excitoxicity, neural stem cell proliferation, and inhibition of stress-induced cdk5 activation. The physiological function of the rapidly degraded p3 peptide is still unclear [51.43] ◀

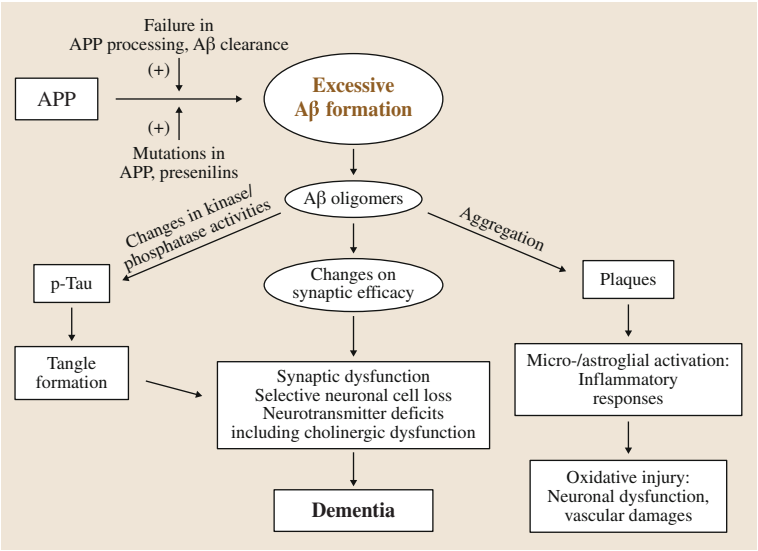

Fig. 51.3 *Amyloid cascade hypothesis of AD: modifying and detrimental effects of Aβ.* The amyloid cascade hypothesis states that the development of AD is initiated by abnormal cleavage of APP, resulting in an imbalance of production and clearance of Aβ. As a consequence, Aβ accumulates in the brain as synaptotoxic oligomers and as fibrillar aggregates in the form of senile plaques, thereby initiating a cascade of pathogenic events that ultimately result in the development of AD [51.47]. For more details, see text. (Graph adapted from [51.48] and [51.10])

The membrane-bound, transmembrane aspartyl protease β-site APP cleaving enzyme 1 (BACE1) has been identified as the β-secretase, while the γ-secretase activity is mediated by a high molecular weight complex consisting of presenilin 1 or 2, nicastrin, anterior pharynx-defective-1 (APH-1), and presenilin-enhancer-2 (PEN-2) (for reviews, see [51.21, 43, 46].

As the clinicopathological features of sporadic AD are indistinguishable from the inherited form of AD, significant efforts have been made to understand the physiological function of the genes tied to FAD. In particular, the development of transgenic mice carrying human FAD genes (for a review, see [51.49]) have essentially contributed to confirm the pathogenic effects of the mutated AD genes and to support the amyloid cascade hypothesis of AD, which has been proposed by [51.48] (Fig. 51.3). Changes in the metabolism of APP and formation and accumulation of Aβ peptides initiates a cascade of events that finally leads to neuronal dysfunction and cell death associated with neurotransmitter deficits and dementia (for reviews, see [51.48, 50]).

51.3.2 Cholinergic Control of APP Processing

The metabolism of APP underlies a strong physiological control with a balanced production and clearance of Aβ. Pathological changes in the processing of APP may favor the β-secretory pathway with the consequence of Aβ accumulation either as synaptotoxic oligomers or as fibrillar aggregates in the form of senile plaques. An abundant number of investigations in the last decades provided compelling evidence that APP processing is controlled by neuronal signaling of several transmitter systems including cholinergic mechanisms. Particularly, G protein-coupled receptors have been disclosed to modify APP processing by affecting the proteolytic activities of the α, β, and γ-secretases (for a comprehensive review, see [51.50]).

This section focuses on the interaction of the cholinergic system and APP processing (for a review, see also [51.50–52]).

Control of APP by mAChR

The first evidence of a link between cholinergic dysfunction and APP processing was provided by observations of a co-localization of acetylcholinesterase (AChE) with Aβ deposits in AD brains [51.53, 54], which was further validated by a number of in vitro studies initiated by [51.55, 56]. Secretion of sAPPα was enhanced by electrical stimulation of tissue slices from rat brains, which could be blocked by the sodium-channel antagonist tetrodotoxin [51.57]. Selective activation of M1/M3-but not M2/M4-mAChR increased sAPPα secretion and decreased total Aβ formation both in vitro [51.58–61] and in vivo in AD patients [51.62, 63]. A similar effect was achieved by direct activation of protein kinase C (PKC) by phorbol ester, indicating that mAChR mediate their effects on APP processing through activation of the phosphatidyl inositol signaling pathway [51.64]. A recent cell culture study provided evidence that PKCα

and PKCε, but not PKCδ, are particularly involved in controlling sAPPα secretion [51.65]. Further studies revealed that other pathways downstream of the mAChR may also be involved in the α-secretase-mediated cleavage of APP, including protein kinase A (PKA), mitogen-activated protein kinase (MAPK), extracellular signal-regulated protein kinase (ERK), tyrosine kinase, and phosphatidylinositol 3-kinase (PI3K) [51.66, 67]. Activation of these signaling cascades shifts APP metabolism towards the α-secretase-mediated pathway with suppressing β-secretase-mediated Aβ formation (for a review, see [51.50]). However, a recent study has questioned the participation of the MEK/ERK pathway in PKC-stimulated and M1-mAChR-dependent formation of sAPPα [51.65].

Selective lesion of basal forebrain cholinergic cells in rat brains [51.68, 69] and administration of selective M1-mAChR agonists to mice [51.70] provided strong in vivo evidence that cortical APP processing is controlled by cholinergic activity originating from the basal forebrain. Scopolamine treatment of transgenic Tg2576 mice resulted in increased levels of fibrillar Aβ, and enhanced α-secretase activity, suggesting that chronic suppression of cortical muscarinic cholinergic transmission may alter the balance between α and β-secretory APP processing by favoring the amyloidogenic route [51.71].

While M2-/M4-mAChR have been observed to inhibit sAPPα release and enhance Aβ production [51.59, 72], studies in AD mouse models provided strong evidence that selective activation of M1-mAChRs drives the APP processing toward the non-amyloidogenic pathway. Thus, administration of the selective M1-mAChR agonist AF267B in the 3xTg-AD model resulted in reduced Aβ and tau pathologies in the hippocampus and cortex [51.73]. The decreased Aβ production indicates an M1-mAChR-mediated shift of APP processing toward the non-amyloidogenic pathway, mediated by an increase in PKC activation, ERK1 and ERK2 phosphorylation, and an increase in α-secretase ADAM17 expression, while the effect of AF267B on tau is obviously due to the reduced GSK3β activity [51.73]. These findings were further supported by genetic ablation studies. Deletion of the M1-mAChR in primary neurons has been shown to increase amyloidogenic APP processing in neurons as evidenced by decreased agonist-regulated shedding of the neuroprotective APP ectodomain sAPPα, and increased production of toxic Aβ peptides. In APP(Swe/Ind) transgenic mice, the loss of M1-mAChRs resulted in increased Aβ level and more amyloid plaques, thus favoring amyloidogenic APP processing [51.72]. This data is further supported by a recent study demonstrating that deletion of M1-mAChR in the 3xTgAD and Tg-SwDI mice increased plaque and tangle levels in the brains of 3xTgAD mice and elevated cerebrovascular deposition of fibrillar Aβ in Tg-SwDI mice, and led to tau hyperphosphorylation, presumably due to changes in the GSK3β and PKC activities [51.74].

Given that α and β-secretory APP processing is differentially regulated through mAChR subtypes, in SH-SY5Y neuroblastoma cells we examined the signaling pathways through which mAChR subtypes control β-secretase activity [51.75]. The expression of the β-secretase BACE1 was found to be differentially regulated in a subtype-specific manner by mAChR. Agonist binding to M1/M3-receptors upregulated BACE1 expression through activation of both PKC and MAPK signaling cascades. In contrast, BACE1 expression was downregulated by activation of M2-mAChR and PKA-mediated pathways [51.75]. These results may partly explain the observed deteriorations of AD patients after initial improvements by AChE-inhibitor or M1-mAChR agonist treatment.

Activated G protein-coupled receptors are desensitized by phosphorylation under the catalytic action of G protein-coupled receptor kinases [51.76]. GRK5, a member of the GRK protein family, is specifically involved in desensitizing activated Gi-coupled M2/M4-mAChR subtypes [51.77]. As M2/M4-mAChRs function primarily as presynaptic autoreceptors on cholinergic neurons, acting as feedback inhibitor of acetylcholine release [51.78], a functional loss of GRK5 should cause prolonged or persistent M2/M4-mAChR signaling leading to reduced acetylcholine release and cholinergic hypofunction. As GRK5 deficiency has been reported in AD [51.79], a role of GRK5 in promoting amyloidogenic APP processing via prolonged activation of M2/M4-muscarinic autoreceptors was suggested [51.80]. Indeed, in AD-like Tg2576 mice lacking one copy of *Grk* gene significantly increased Aβ accumulation including enhanced plaque load, and sAPPβ production was observed, which could be reversed by the administration of selective M2-mAChR antagonists [51.80].

As Aβ has been shown to be one of the main causes of the functional deficiency of GRK5 in AD [51.79], a positive feedback loop among Aβ, GRK5 deficiency, and cholinergic dysfunction has been postulated, by which each component can mutually promote each other, thus exacerbating both amyloid pathology and cholinergic dysfunction [51.80].

Control of APP by nAChR

Nicotine through action on nAChR has also been observed to modulate APP processing both in cell culture [51.81–84] and in vivo [51.85] by favoring the non-amyloidogenic pathway when treated with low doses of nicotine [51.86].

To reveal which subtype of neuronal nAChRs mediates the actions of nicotine on APP processing, subtype-selective suppression/transfection studies were performed in cell line cultures. Suppression of the α7-nAChR gene by siRNA indicated that α7-nAChR stimulation increases sAPPα formation by enhanced α-secretase activity, accompanied by activation of MAPK signaling, and improving antioxidant defenses [51.84].

By using cell lines expressing different subtypes of nAChRs, it has been detected that stimulation of both α4- and α7-nAChR by nicotinic compounds is involved in driving the APP processing toward the non-amyloidogenic pathway by increasing sAPPα secretion and attenuating Aβ production [51.87]. While activation of α7 nAChR by nicotine and specific α7-nAChR agonists promotes non-amyloidogenic APP processing by decreasing the γ-secretase activity with no effects on α-secretase and β-secretase activity [51.88], the decrease in Aβ production by activation of α4β2 nAChR is mediated through regulation of BACE1 transcription, presumably via the ERK1/NFκB pathways [51.89].

These findings have also been validated by in vivo studies. Chronic treatment of transgenic Tg2576 APP mice with nicotine significantly reduced Aβ plaque deposition as compared to non-treated controls [51.90]. In a rat model of AD chronic nicotine was observed to restore normal Aβ levels and prevented short-term memory and LTP impairment [51.91]. Interestingly, tobacco-smoking elderly people demonstrated less Aβ deposition in the entorhinal cortex as compared to non-smokers [51.92].

51.4 Modulation of Cholinergic Function by Aβ

While impaired cholinergic neuronal signaling may induce pathologically enhanced Aβ production, there is also abundant evidence of the cytotoxic, modifying, and suppressing capacities of Aβ on cholinergic cells and function, suggesting the generation of vicious feedback loops. This section reviews the effect of Aβ on viability and signaling of cholinergic neurons including NGF signaling in cholinergic cells.

51.4.1 Cytotoxicity of Aβ on Cholinergic Cells

The most predominant Aβ peptides detected in Alzheimer plaques are Aβ (1–40) and Aβ (1–42). Aβ (1–42) is more fibrillogenic and displays higher neurotoxicity in vivo than Aβ (1–40) [51.93]. The insoluble, high-molecular weight fibrils are major components of the senile plaques in the Alzheimer brain and have been assumed for a long time to be the most toxic forms responsible for cholinergic neurodegeneration (see, e.g., [51.35, 94]). However, recent evidence indicates that soluble oligomers of Aβ (such as low-molecular weight monomers, oligomers and amyloid-derived diffusible ligands (ADDLs)), as well as protofibrils represent the main neurotoxic species leading to early neuronal dysfunction and memory deficits in AD (for reviews, see e.g., [51.95, 96]). In different cell and animal models prefibrillar assemblies of Aβ have been shown to induce neurotoxicity (for reviews, see e.g., [51.51, 97], electrophysiological changes [51.98], modulation of synaptic plasticity [51.98, 99], and disruption of cognitive function [51.100, 101], which may explain why early onset of cholinergic dysfunction is in progress before there is considerable plaque formation in AD. Indeed, in AD brains, it has been observed that the severity of neurodegeneration correlates best with the pool of soluble Aβ rather than with the number of insoluble Aβ plaques [51.102]. This data fits well with observations in AD-like transgenic mouse models revealing decreases in cholinergic fibre density, in mAChR and nAChR binding levels in the cerebral cortex already before onset of plaque deposition [51.103–109]. Studies in BACE1 knock-out mice that do not produce any significant amount of Aβ [51.110] provided further evidence of Aβ-induced cholinotoxicity. BACE1 gene deletion rescued memory deficits and cholinergic dysfunction in AD mice which was associated with tremendously reduced Aβ levels [51.111].

In a recent study on brain autopsy in AD patients, the relationship between various Aβ oligomer assemblies found in AD brains with the levels of fibrillar Aβ and cholinergic synaptic function was examined, suggesting that only distinct Aβ oligomers induce impairment of cholinergic neurotransmission in AD [51.112].

51.4.2 Modulation of Cholinergic Transmission by Subtoxic Aβ

Aβ is also produced under normal conditions and secreted in the brain as a soluble peptide [51.113, 114], which raised the possibility that Aβ may also play a physiological role. This is supported by cell culture studies providing evidence of a modulatory role of soluble Aβ at subtoxic concentrations on cholinergic neurotransmission [51.35]. Soluble Aβ at pM to nM concentrations strongly inhibited the potassium-stimulated release of acetylcholine from hippocampal slices and reduced the high-affinity uptake of choline in synaptosomal preparations from cortex and hippocampus [51.115]. Choline deprivation has been suggested to render basal forebrain cholinergic neurons particularly vulnerable [51.116] and may initiate autocannibalism of cholinergic cells by cleavage of membrane phosphatidylcholine to replenish the choline availability [51.35].

Aβ peptides at subtoxic concentration also decreased the intracellular acetylcholine concentration in primary cultures [51.117] and resulted in reduced activity of ChAT but not AChE activity in cholinergic SN56 cells [51.118]. As exposure of primary neurons by Aβ reduced glucose uptake, the Aβ-mediated decline in intracellular acetylcholine might be due to the limited supply of acetyl-CoA, a prerequisite to acetylcholine synthesis.

AChE has been found in amyloid plaques. The association of AChE and Aβ alters the enzymatic properties of AChE and renders Aβ more toxic by accelerating assembly of Aβ into fibrils (for a review, see [51.119]).

Aβ at nanomolar concentration did not affect mAChR ligand binding, but impaired M1-mAChR associated signaling in primary septal and cortical cultures, which resulted in reduced inositol phosphate production and decreased calcium release from the intracellular pool [51.120].

Aβ exposure of primary cultures and brain slices have also been found to affect nAChR binding and signaling, but depending on the concentration of Aβ (see also Sect. 51.4.4).

In conclusion, in vitro data indicate that Aβ at physiological concentration (nM to pM) appears to play a potent negative modulator of acetylcholine synthesis and release, and interferes with normal cholinergic signaling mediated through mAChR and nAChR subtypes.

However, APP itself may also affect cholinergic transmission. APP was found to interact with the high-affinity choline uptake by mediating presynaptic localization and activity of the high-affinity choline transporter [51.121].

51.4.3 Effect of Aβ on mAChR

In AD brains the number of M1-mAChR has been reported to be mostly unaffected, while in some studies a decreased number of the M2-mAChR subtype was described as associated with the impaired coupling of the mAChR to heterotrimeric GTP-binding proteins (G proteins) [51.36].

While in vitro studies did not provide any evidence of a direct interaction of Aβ with mAChR binding sites, pathologically accumulating Aβ appears to affect mAChR signaling mainly by influencing downstream events, finally leading to loss of mAChRs and cholinergic dysfunction [51.103, 107, 109, 122]. Recently, a mechanism was proposed to explain how Aβ may interfere in mAChR signaling by interruption of the mAChR G-protein coupling [51.123].

Aβ accumulation triggers increased generation of reactive oxygen species inducing dimerization of the angiotensin type 2 receptor, which can further cross-link and lead to oligomerization of the angiotensin type 2 receptor dimers. The angiotensin type 2 oligomers sequester the G-protein $G\alpha_{,q/11}$, thus preventing the coupling of $G\alpha_{,q/11}$ to the M1-mAChR. Dysfunctional coupling of M1-mAChR and $G\alpha_{,q/11}$ is considered to mediate cholinergic deficits and cell loss, tau phosphorylation, and memory impairments [51.123].

Recently, in transgenic AD-like Tg2576 mice, Aβ-dependent inactivation of the JAK2/STAT3 axis was reported. This resulted in downregulation of ChAT activity and desensitization of the M1-mAChR, thus providing evidence of another mechanism of how enhanced level of Aβ can impair mAChR signaling and induce cholinergic dysfunction [51.124].

51.4.4 Effect of Aβ on nAChR

AD patients show a significant reduction in nAChRs, which has been documented by Western blotting [51.125, 126], immunohistochemical analysis [51.127, 128], or radioligand binding studies ([51.129]; for reviews, see [51.130, 131]). The most vulnerable neurons appear to be those expressing high levels of α7-nAChR [51.132]. As α7-type AChRs are highly expressed in brain regions relevant to memory functions, it has been suggested that α7 AChR has an important role in the development of AD (for a review, see [51.133]).

Exposure of PC12 cells to Aβ resulted in a significant decrease in nAChR, which led to the suggestion that Aβ can damage nAChR [51.134, 135]. Similarly, in rats chronically infused with Aβ peptides, significant decreases in the levels of nAChR subunits α7, α4, and β2, accompanied by an increased BACE expression, have been observed [51.136]. Both actions of Aβ could be reversed by chronic delivery of nicotine, suggesting inhibitory rather than damaging effects of Aβ on nAChRs [51.136].

On the other hand, there are reports that Aβ may affect nAChR by direct binding with high-affinity to nAChR, in particular to the α7 subtype [51.132, 137–140]. Other studies have been unable to confirm these findings [51.135, 141]. Aβ has been observed to act as an agonist of α7-nAChR, mediating the activation of the ERK2 MAP kinase signaling cascade [51.142]. Other groups have reported inhibitory actions of Aβ on α7 nAChR [51.143], which appears to depend on the concentration of Aβ; low concentration can activate, higher concentrations desensitize α7 nAChR [51.144]. This compares well with observations in triple transgenic mice overexpressing mutated human APP, presenilin-1 and tau (3xTg-AD), which demonstrate an age-dependent reduction in α7-nAChRs as compared to age-matched non-transgenic mice. The loss of α7-nAChRs is preceded by intracellular Aβ accumulation and is restricted to brain regions that develop Aβ pathology [51.139], a finding which mimics the situation detectable in brains of AD patients. Therefore, parts of the cholinergic deficits produced in Alzheimer's disease as well as in transgenic APP mice could be attributed to the suppression of cholinergic functions by Aβ peptides.

Recently, it was hypothesized that α7-nAChR may represent the missing link in understanding AD etiopathology [51.145]. This hypothesis stresses recent experimental findings that nicotinic agonists and Aβ may compete for the α7-nAChR binding site, whereas intracellular signaling cascades are activated that control either survival or cell death pathways, respectively [51.130]. Moreover, a recent study supports α7-nAChR as a mediator of Aβ-induced pathology in AD by demonstrating that both agonists and antagonists may modulate Aβ-induced tau phosphorylation through GSK-3β [51.146]. Deletion of the α7-nAChR gene in the PDAPP Alzheimer mouse model improved cognitive deficits and synaptic pathology, suggesting that blocking the α7-nAChR function may alleviate symptoms of AD and may represent a potential treatment strategy [51.147].

Recently, the functional effects of Aβ fibrils and oligomers on nAChRs were examined by measuring intracellular calcium levels in neuronal cells [51.148]. It was demonstrated that fibrillar Aβ exerts neurotoxic effects mediated partly through a blockade of α7-nAChRs, whereas oligomeric Aβ may act as a ligand activating α7-nAChRs, thereby stimulating downstream signaling pathways [51.148].

On the other hand, overexpression of APP (SWE) gene has been shown to influence the expression of nAChRs and resulted in neurotoxicity [51.149], while the disruption of cholesterol homeostasis as observed in AD brain may affect cholinergic synapses by decreasing the number of nAChR (see, e.g., [51.133]).

51.4.5 Aβ and Neurotrophin Signaling

For their maintenance and survival basal forebrain cholinergic cells require the neurotrophic support by the nerve growth factor (NGF) that is produced and released by cholinoceptive cells in the cortex. NGF exerts its action by binding to the high-affinity NGF-specific receptor tyrosine kinase, trkA, and the low-affinity, pan-neurotrophin receptor, p75NTR, which are expressed by basal forebrain cholinergic neurons, while neurons in other brain regions express little or no p75NTR. Receptor-bound NGF is internalized and retrogradely transported to the nucleus of basal forebrain cholinergic cells (for a review, see [51.150]). Transgenic mice that have been modified to produce anti-NGF antibodies exhibit degeneration of basal forebrain cholinergic cells and partly mimic AD-like pathology, suggesting that NGF has an important role for cholinergic cell viability [51.151].

While NGF expression is not altered in AD, dysfunction of cytoskeletal transport including reduced axonal transport of NGF has been assumed to play an important role in the development of AD pathology [51.150, 151], as both APP and tau are involved in axonal transport. Increased doses of APP markedly decreased retrograde transport of NGF and resulted in degeneration of forebrain cholinergic neurons [51.152].

Trophic deprivation has been observed to affect the cleavage of surface APP by BACE1, releasing sAPPβ, which is further cleaved to N-APP, which may bind to the death receptor 6 (DR6) to trigger degeneration [51.44]. On the other hand, APP may directly interact with p75NTR, which mediates death of basal forebrain cholinergic neurons [51.153].

Recently, it was discovered that the precursor of NGF, the proNGF, also displays biological activities,

but that these activities are distinct from that of mature NGF ([51.17]; for a review, see [51.154]). proNGF may interact with sortilin as co-receptor for p75NTR and induce p75NTR-dependent apoptosis or may, to a lesser extent, bind to trkA and mediate trA-dependent neuronal survival [51.155]. In AD brains a reduced conversion of proNGF into mature NGF, and an increased NGF degradation has been observed [51.156], suggesting an imbalance of NGF/proNGF signaling in AD that may contribute to the cholinergic cell loss.

Because of the select expression of p75NTR by basal forebrain cholinergic neurons, a role of p75NTR in cholinergic dysfunctions in AD has been suggested (for a review, see [51.157]). There is abundant evidence from in vitro studies that p75NTR increases the susceptibility of cells to Aβ toxicity, suggesting that p75NTR mediates Aβ cytotoxicity by direct interaction with Aβ, or acting on signal transduction pathways mediated by p75NTR [51.158, 159]. These observations were further confirmed recently by a number of in vivo studies. Aβ injected into the hippocampus of p75NTR/knock-out mice caused less neuronal death as compared to that observed in wild type (p75NTR+/+) mice [51.160]. Transgenic Thy1-hAPPLondon/Swe Alzheimer-like mice lacking wild type p75NTR did not display Aβ-associated basal forebrain cholinergic neuritic dystrophy and reduced cholinergic cortical fibre density as has been observed in transgenic Thy1-hAPPLondon/Swe mice with intact p75NTR [51.161]. The data provide evidence that p75NTR may be a major player in Aβ-associated cholinergic deficits, mediated through a p75NTR-activated c-Jun N-terminal kinase pathway [51.162].

Using another genetic approach (crossing the p75NTR knock-out mouse with the transgenic APPswe/PS1dE9 AD mouse), p75NTR has been shown to regulate Aβ deposition by increasing neuronal Aβ production [51.163]. The extracellular domain of p75NTR after shedding from the membrane (by an α-secretase activity) may bind and sequester Aβ and thus inhibit Aβ aggregation and reduce Aβ deposition. Moreover, it may also block the interaction of p75NTR and Aβ or proNGF by competitive binding, thus attenuating the p75NTR-dependent neurotoxicity and cell death [51.163]. On the other hand, p75NTR has been shown to be upregulated in two strains of AD transgenic mice compared to corresponding wild-type mice, which correlated with the age-dependent accumulation of Aβ (1–42) level [51.164]. As activation of p75NTR signaling was observed to stimulate Aβ production [51.163], the Aβ-induced p75NTR upregulation may initiate a vicious cycle that accelerates AD development [51.164].

51.5 Tau and Cholinergic Transmission

While no tau mutations have been described in the brains of AD patients, pathogenic mutations in the tau genes cause frontotemporal dementia [51.165], suggesting that posttranscriptional alterations in tau gene expression may contribute to the cognitive deficits in AD presumably also by interacting with the cholinergic transmission. Several studies have demonstrated that activation of nAChR results in a significant increase in tau phosphorylation, whereas mAChR activation may prevent tau phosphorylation (for reviews, see [51.130, 145]. Nicotine was found to induce tau phosphorylation at those sites that were also hyperphosphorylated in AD, presumably mediated through activation of the α7 subtype of nAChR [51.143]. This was further emphasized by observations in triple transgenic 3xTg-AD mice that develop age and regionally-dependent accumulation of both plaques and tangles as well as progressive deficits in cognition [51.166–168]. Chronic nicotine administration to one-month-old 3xTg-AD mice for five months did not change soluble Aβ levels but resulted in a striking increase in phosphorylation and aggregation of tau, which appeared to be mediated by p38-MAP kinase [51.139].

Cholinergic basal forebrain neurons have been shown to demonstrate tau pathology both in patients with mild cognitive impairment and in AD patients. Single cell gene expression profiling in individual human cholinergic basal forebrain neurons revealed a shift in the ratio of three-tandem repeat tau to four-tandem repeat tau during the progression of AD but not during normal aging [51.169].

Basal forebrain pretangles and tangles have been observed prior to the pathology in the entorhinal/perirhinal cortex, indicating that abnormalities in cortical cholinergic axons and tau pathology within the basal forebrain cholinergic system occur very early in the course of life and increase in frequency in old age and AD [51.170, 171]. In a tau transgenic mouse model, tau pathology has been observed in both hippocampus and basal forebrain, which was associated with a significant reduction in medial septal cholinergic neurons

and a decreased uptake and retrograde transport of NGF by cholinergic nerve terminals [51.172], suggesting that tau pathology may participate in cholinergic degeneration [51.172].

In conclusion, as Aβ may trigger tau phosphorylation, and Aβ pathology precedes tau pathology, tau-mediated changes in cholinergic cells may represent an indirect effect of pathogenic Aβ accumulation.

51.6 Neurovascular Function in AD

51.6.1 Cerebrovascular Abnormalities and Dysfunction in AD

In the brain, the cerebral blood flow is tightly regulated to assure adequate and timely blood supply to brain regions that have momentarily high energy demand because of enhanced neural activity, a phenomenon called functional hyperemia [51.174]. Already at very early stages of AD, changes in the cerebral blood flow, such as reduced blood supply at rest and altered perfusion to activated areas have been observed, which provided evidence to suggest a causal relationship between vascular mechanisms and the development of sporadic AD. This vascular hypothesis of AD was first formulated by [51.175] (for reviews, see [51.173, 176, 177], Fig. 51.4). Insufficient cerebral blood flow may induce hypoxia-sensitive pathways leading to inflammation with upregulation of pro-inflammatory cytokines and to oxidative stress with generation of reactive oxygen species, which may be detrimental to

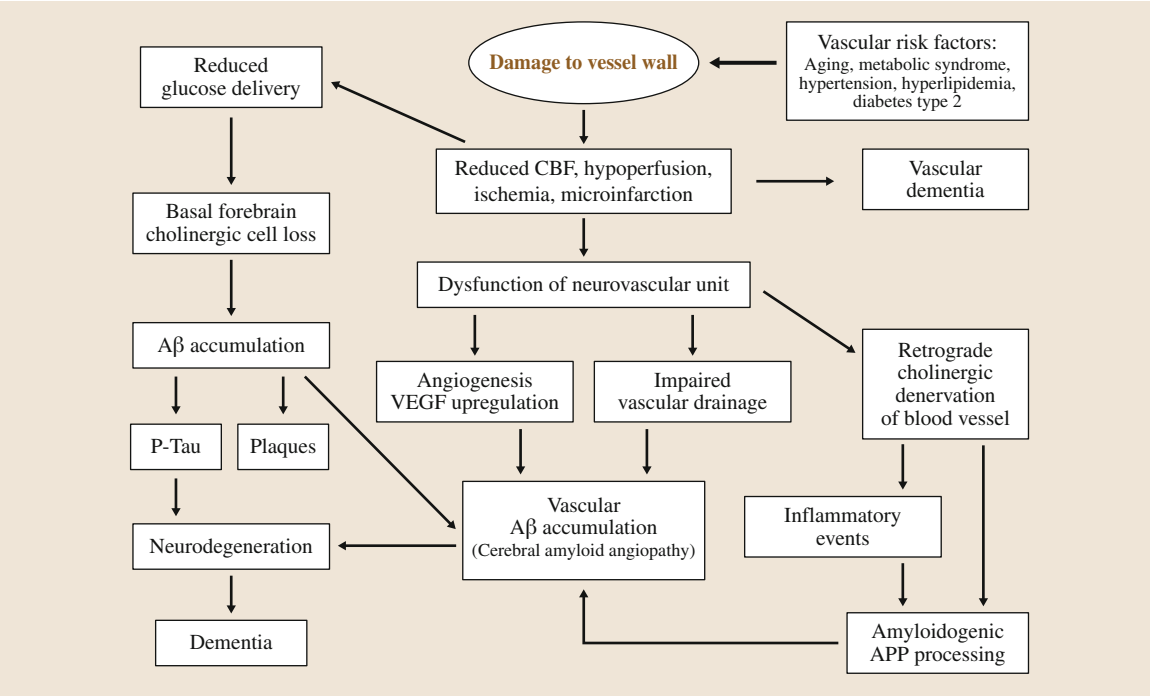

Fig. 51.4 *The vascular hypothesis of AD.* The vascular hypothesis for AD is based on findings that vascular risk factors damage the brain microvasculature, which results in chronic hypoperfusion, reduced cerebral blood flow, and reduced glucose supply [51.173]. Vascular synaptic damage may induce retrograde cell death of cholinergic neurons, which may trigger inflammation, favor amyloidogenic APP processing and cerebral amyloid angiopathy (CAA), and initiate tau pathology. Damage to vascular endothelial cells may induce angiogenesis and VEGF upregulation, which further promotes β-amyloidogenesis. As basal forebrain cholinergic neurons particularly respond sensitively to reduced glucose supply by synaptic dysfunction and degeneration, the cascade of events leading to the development of AD is further exacerbated. For details, see text

vascular integrity and function. Indeed, cerebrovascular abnormalities such as thickening of the microvascular basement membranes, decreased luminal diameter, and microvascular degeneration, in particular in the temporal-parietal cortex, have frequently been observed in AD patients [51.177].

In advanced AD cases Aβ deposition occurs also in cerebral vessels (cerebral amyloid angiopathy CAA), which may result in smooth muscle degeneration and weakening of the vascular wall, impairing vasomotor function, and increasing the risk of cerebral hemorrhage [51.178]. Indeed, it has been suggested that the recently detected brain microbleeds (small dot-like lesions) in AD brains are the missing link between the amyloid cascade hypothesis and the vascular hypothesis [51.179]. The CAA observed during normal aging and in the majority of AD cases is likely to be caused by the failure of Aβ elimination from the brain parenchyme. It has been suggested that receptor for advanced glycation end products (RAGE), and low density lipoprotein receptor related protein 1 (LRP1) play a role in controlling the Aβ transport through the blood brain barrier (for a review, see [51.180]). A recent study in transgenic AD-like mice (Tg2576) further supported the *clearance hypothesis* demonstrating impaired perivascular solute drainage from the brain in aged mice as compared to younger ones [51.181]. Currently, in a transgenic AD mouse model with strong CAA pathology, it has been suggested that early perivascular astrocytic dysfunction play a particular role in impairing cerebrovascular and metabolic pathology [51.182].

Studies in transgenic mouse models of AD suggested that the compromised cerebral hemodynamics observed in AD appears to be associated with inflammation [51.183]. The local activation of microglia and reactive astrocytes is accompanied by production and secretion of proinflammatory cytokines such as interleukin-1β (IL-1β), tumor necrosis factor (TNF)-α, and transforming growth factor (TGF)-β1. Endothelial cells are known to respond sensitively to inflammatory stimuli by production of reactive oxygen species that may further exacerbate the vascular damages [51.184].

A number of risk factors are assumed to mediate the cerebrovascular dysfunctions and to trigger AD pathology, such as hypertension, hyperlipidemia, enhanced homocysteine levels, diabetes type 2, metabolic syndrome (obesity, hypertension, cardiovascular disease, atherosclerosis), as well as genetic factors (ε4 allele of ApoE), age, and life style [51.173, 185, 186]. However, it is still a matter of debate whether neurovascular dysfunction and vascular lesions play a causative role for the neurodegenerative processes as suggested by a number of reports (for a review, see [51.180]). However, regardless of that, cerebrovascular diseases appear to play an important role in determining the presence and severity of the clinical symptoms of AD [51.187].

51.6.2 Effect of Aβ on Brain Vascular System

The microvascular degenerations observed in AD may also be the consequence of the vasoactive detrimental effects of Aβ. Aβ is a potent vasoconstrictor in the brain, as has been shown in vivo and in vitro by application of exogenous Aβ to normal blood vessels and to mouse cortices. On the other hand, Aβ may cause degeneration of both the larger perforating arterial vessels as well as cerebral capillaries, presumably mediated through the induction of reactive oxygen species by activation of NADPH oxidase, which may subsequently severely affect regulation of cerebral blood vessels and brain perfusion, as well as impair the blood brain barrier (for reviews, see [51.174, 178, 188, 189]). Indeed, observations in transgenic AD-like mice revealed Aβ-mediated impairments of endothelium-dependent regulation of cortical microcirculation, abnormal vascular autoregulation, reduced cerebral blood flow, and attenuated cerebrovascular reactivity to functional hyperemia already before onset of any plaque load, further supporting the link of Aβ to the mechanisms of vascular dysfunction (for comprehensive reviews, see [51.174, 177]. However, the view that elevated soluble Aβ levels are sufficient to cause cerebrovascular dysfunction has also been challenged by studies on the Tg2576 transgenic mouse model [51.190].

Moreover, Aβ peptides have been described to inhibit angiogenesis both in vitro and in vivo [51.191, 192], and deregulation of angiogenic factors may contribute to various neurological disorders including neurodegeneration (for a review, see [51.193]). One of the key angiogenic factors, the vascular endothelial growth factor (VEGF), a highly conserved heparin-binding protein [51.194], was originally found in vascular endothelial cells and is able to induce vascular endothelial cell proliferation, migration, and vasopermeability in many types of tissue [51.195].

Increased intrathecal levels of VEGF have also been observed in brains of AD patients as compared to age-matched healthy individuals [51.196–198], which has been correlated with the clinical severity of the disease [51.199]. However, the functional significance of VEGF upregulation in the pathogenesis and progres-

sion of AD is still a matter of debate. While VEGF and other angiogenic factors were found to be enhanced in AD [51.200–203], neovascularization has been observed only in the hippocampus of AD patients [51.203].

51.6.3 Effect of Ischemia and Hypoperfusion on APP Processing

There are reports that ischemia and hypoperfusion may trigger accumulation and cleavage of APP into Aβ, and its deposition in the brain, as well as hyperphosphorylation of tau and PHF formation (for a review, see [51.180]). The upregulation of VEGF in response to hypoxic, ischemic, or hypoglycemic stress [51.204–206], suggests its involvement also in the processing of APP. In turn, APP is also highly expressed in the endothelium of neoforming vessels [51.207], and inhibitors of β- and γ-secretases have been reported to inhibit angiogenesis and tumor growth [51.207], suggesting a role of APP metabolism also during angiogenesis. Recently, VEGF has been shown to also be involved in the induction of microglial-mediated inflammation by Aβ deposits via the microglial VEGF receptor subtype Flt-1 serving as a chemotactic receptor to mobilize microglial cells [51.199].

As vascular endothelial cells are also capable of expressing and to secreting APP [51.208], it has been hypothesized that VEGF may also be involved in the formation and deposition of Aβ. This hypothesis has been addressed by examining the effect of VEGF on APP processing in brain slice cultures, as well as in primary neuronal, astroglial, and vascular endothelial cells derived from AD-like transgenic Tg2576 mice [51.209]. The exposure of brain slices by VEGF resulted in an inhibition of the formation of soluble Aβ peptides, which was accompanied by a transient decrease in β-secretase activity, as compared to controls [51.209]. Similar studies in primary neurons, astrocytes, and endothelial cells expressing the Swedish mutation of human APP, have further provided evidence that VEGF affects APP processing but differentially acts in cells that form the neuron-glia-vascular unit [51.210].

51.6.4 Effect of Aβ on Cholinergic Function in the Brain Vascular System

There is a large body of evidence that cerebral blood flow and local glucose delivery is controlled by neuronal activity, known as neurometabolic and neurovascular coupling [51.177, 184, 211, 212]. Dysfunctions of the regulation of the cerebral blood circulation may affect vital control mechanisms that ensure delivery of adequate amounts of substrate and maintain the homeostasis of the microenvironment of the neurovascular unit. The neurovascular unit defines the cellular interaction between brain capillary endothelial cells, the end feet of perivascular astrocytes, and neuronal axons [51.184]. Pial arteries at the surface of the brain are densely innervated by perivascular nerves that originate from autonomic and sensory ganglia, whereas intracerebral arterioles and capillaries receive afferents that originate from subcortical neuronal centers as well as from local cortical interneurons (for a comprehensive review, see [51.213]).

Based on findings in immunocytochemistry and electron microscopy, it is known that the cholinergic axons originating from the basal forebrain project not only to the cortical neuropile but also to arterioles, capillaries, and to perivascular astrocytes within the cerebral cortex [51.214]. Furthermore, there is physiological evidence that the central cholinergic pathways are involved in the regulation of cerebral cortical blood flow. Electrical or chemical stimulation of cholinergic basal forebrain neurons results in increased cerebral blood flow [51.215]. The involvement of ACh as a neurotransmitter in the control of regional cerebral blood flow has been further demonstrated by administration of cholinergic drugs [51.216]. While application mAChR antagonist decreased cerebral blood flow, the inhibition of AChE led to increased cerebral blood flow [51.217]. This response was found to be dependent on nitric oxide (NO) production and presumably to be mediated through the M5-mAChR subtype [51.216]. The basal forebrain cholinergic fibers can either directly affect the cerebrocortical microvasculature or innervate subpopulations of GABAergic interneurons releasing the vasodilators NO and VIP [51.218]. The interneurons appear to serve as a functional relay to adapt perfusion to locally increased neuronal activity [51.212].

On the other hand, damage to the neurovascular unit either by oxidative stress, inflammation, or Aβ accumulation may induce degeneration of vascular cholinergic nerve terminals and subsequent retrograde cell death of basal forebrain cholinergic neurons. The loss of cholinergic innervation of components of the neurovascular unit may affect APP processing with enhanced Aβ formation and deposition, microglia activation, and inflammation, thus suggesting a link between Aβ production, impairments in cerebrovascular function, and basal forebrain cholinergic deficits in AD (cholinergic-vascular hypothesis of AD; [51.219, 220]). Indeed, a semiquantitative immunohistochemical

study on aged Tg2576 mice revealed an Aβ-mediated decrease in cholinergic innervation of cortical blood vessels [51.221], which has been assumed to contribute to the alterations of the cerebrovascular system observed in transgenic Tg2576 mice [51.209].

51.6.5 Effect of Glucose Deprivation on Cholinergic Neurons

AD patients feature decreased basal cerebral glucose utilization, presumably as a consequence of the cerebrovascular dysfunction and compromised cerebral hemodynamics [51.177]. Glucose deprivation has been suggested to render basal forebrain cholinergic neurons particularly vulnerable [51.222], as they require glucose not only for energy production but also to synthesize acetylcholine from choline and acetyl-CoA, which is generated by glucose degradation through glycolysis. As in cholinergic neurons energy production and acetylcholine synthesis compete for acetyl-CoA; reduced glucose supply with the consequence of a decline in acetyl-CoA synthesis may easily evoke energy deficits, leading to impairment of their function and structural integrity, as neurons have no energy reserves [51.222]. Interestingly, it has been proposed that these early abnormalities in brain glucose and energy metabolism in AD are caused by an *insulin-resistant brain state*, which appears to be responsible for the increased Aβ accumulation, tau phosphorylation, and cognitive deficits (for a review, see [51.223]).

On the other hand, reduced glucose utilization in AD may also be caused by progressive Aβ accumulation affecting expression and/or activity of key enzymes of the glycolysis in brain cells. In a mouse model of AD (Tg2576 mice) expression and activity of the phosphofructokinase (PFK), a key enzyme in regulation of glycolysis, was studied [51.224]. In a 24-month-old transgenic Tg2576 mouse cortex, but not in 7, 13, or 17-month-old mice, the copy number of PFK-C mRNA, the PFK protein level and PFK enzyme activity was significantly reduced as compared to non-transgenic litter mates, while the mRNA level of the other PFK isoforms did not differ between transgenic and non-transgenic tissue samples. In situ hybridization in brain sections from aged Tg2576 mice revealed reduced PFK-C mRNA expression in Aβ plaque-associated neurons and upregulation in reactive astrocytes surrounding Aβ deposits. The data demonstrate that long-lasting high Aβ burden impairs cerebral cortical glucose metabolism by reducing PFK activity in Aβ plaque-associated neurons and concomitant upregulation in reactive, plaque-surrounding astrocytes [51.224]. Therefore, Aβ-induced alterations in glucose metabolism may contribute to cholinergic dysfunction and cell loss.

In conclusion, the basal forebrain cholinergic system plays a significant role in neurovascular regulation and blood flow control, and vascular cholinergic deficits caused by damaged microvasculature may vice versa contribute to amyloidogenic APP metabolism, thus exacerbating AD pathology.

51.7 Interrelationship of Amyloid, Cholinergic Dysfunction, and Brain Vascular Impairments

For a variety of pathological features that have been attributed to play major roles in triggering AD, three major hypotheses of AD have been stressed.

51.7.1 Cholinergic Hypothesis of AD

The early occurrence of basal forebrain cholinergic cell loss in FAD and in advanced stages of sporadic AD, as well as the correlation of clinical dementia ratings with the impairments of cholinergic function have suggested a role of the cholinergic system in the pathogenesis of AD (Fig. 51.5). Indeed, both in vitro and in vivo studies provided evidence of a modulatory role of cholinergic signaling on APP metabolism, and impairments in mAChR and nAChR signaling may drive the APP processing toward the amyloidogenic path and thus contribute to the amyloid pathology of AD.

Selective activation of M1/M3 but not M2/M4-muscarinic acetylcholine receptors (mAChR) increased sAPPα secretion and decreased total Aβ formation, mediated through activation of PKC and/or MAPK signaling. Agonist action on nAChR has been shown to modulate APP processing by favoring the non-amyloidogenic pathway and to inhibit β-amyloid fibril formation. AChE was found to promote the aggregation of Aβ and plaque formation by forming a complex with the growing fibrils. Vice versa, Aβ may increase AChE around Aβ plaques through the action of Aβ on α7-

nAChR. At nanomolar concentrations soluble Aβ has been observed to inhibit markers of cholinergic function. Activation of α7-nAChR may induce degradation and clearance of Aβ at least in transgenic APPswe mice.

NGF plays a maintaining role for cholinergic cells, but was also observed to modulate cholinergic control of APP processing and to influence APP metabolism by favoring the non-amyloidogenic pathway through TrkA receptors, but to increase neuronal Aβ production via p75NTR.

Cholinergic dysfunctions may also be caused by impaired glucose utilization, damages of the microvasculature (retrograde cholinergic terminal degradation), and impaired NGF signaling (Fig. 51.5).

51.7.2 Amyloid Cascade Hypothesis

The discovery of Aβ as the major component of senile plaques, one of the histopathological hallmarks of AD, provided the basis for the establishment of the amyloid cascade hypothesis of AD, stating that Aβ accumulation is the essential event leading to neuronal dysfunction and cell death associated with neurotransmitter deficits and dementia (Fig. 51.3). This hypothesis was particularly supported by investigations of transgenic mice carrying human FAD genes. These mice mimic some of the key features of AD such as amyloid plaques, cognitive deficits, and impairments in neuronal signaling and microvasculature.

Both in vitro and in vivo studies demonstrated the detrimental and modifying effects of accumulating Aβ on cholinergic synaptic events by blocking nAChR, impairing mAChR signaling, inhibiting AChE, and interfering with NGF signaling by binding to p75NTR.

Aβ may damage vascular endothelial cells and demonstrates vasoactive properties by impairing perfusion, perivascular drainage, and cerebral blood flow, as well as inhibiting angiogenesis. It may trigger inflammation by activation of microglial and reactive astroglial cells, which secrete pro-inflammatory cytokines (IL-1β, TNFα, TGFβ), and mediates tau pathology, which finally leads to neurodegeneration and dementia. IL-1β and TNFα have been shown to selectively degenerate cholinergic basal forebrain cells, while IL-1β has been found to upregulate APP expression and to stimulate the amyloidogenic route of APP processing. Furthermore, IL-1β may also promote activity and expression of AChE.

51.7.3 Vascular Hypothesis of AD

The vascular hypothesis for AD is based on findings that vascular risk factors damage the neurovascular unit, which results in chronic hypoperfusion, reduced cerebral blood flow, and reduced glucose supply (Fig. 51.4). As brain capillary endothelial cells receive a cholinergic input from the basal forebrain, the vascular synaptic damages may induce retrograde cell death of cholinergic neurons, which may trigger inflammation and

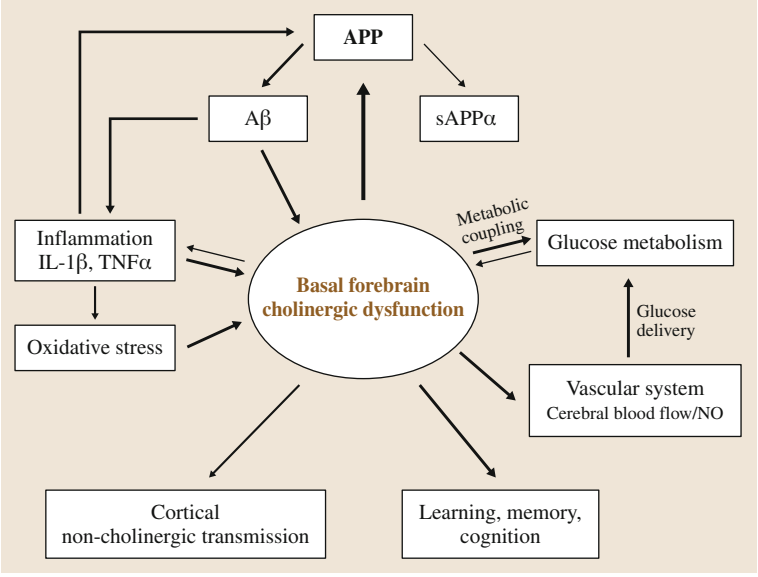

Fig. 51.5 *The cholinergic hypothesis of AD.* The cholinergic hypothesis states that a dysfunction of basal forebrain cholinergic neurons causes the cognitive decline observed in AD patients, which is based on experimental findings that cholinergic neurotransmission has a fundamental role in learning and memory [51.39]. The hypothesis has been further validated by experimental observations that APP processing is controlled by cholinergic neuronal activity. Cholinergic dysfunction shifts the route of APP processing toward the amyloidogenic route, which again contributes to cholinergic deficits. Cholinergic dysfunctions may also be caused by impaired glucose utilization, damage to the microvasculature (retrograde cholinergic terminal degradation), and impaired NGF signaling (for details, see text)

alterations in neurotrophin signaling, may favor amyloidogenic APP processing including CAA, and initiate tau pathology. Damage to vascular endothelial cells may induce angiogenesis and VEGF upregulation, which further promotes β-amyloidogenesis. As basal forebrain cholinergic neurons particularly respond very sensitively to a reduced glucose supply by synaptic dysfunction and degeneration, the cascade of events leading to the development of AD is further exacerbated (Fig. 51.4).

51.8 Computational Modeling of the AD Brain

An understanding the pathogenesis of AD has been achieved by addressing a number of different approaches, ranging from investigations of the role and function of key proteins of AD pathology, exploring specific mechanisms and signaling cascades, neurophysiological and highly sophisticated neuroimaging techniques, up to the application of recently developed high-performance techniques of gene expression profiling and proteomic analyses, as well as genome-wide association studies. Consequently, during the last decades of research in neuroscience a vast number of experimental data and datasets have been accumulated and collected in various databases for genes, proteins, molecule structures, enzyme kinetics, etc., which require great computational effort and IT technology to be efficiently handled. On the other hand, computational approaches may provide useful tools not only to handle the abundant data available so far, but also to allow us to construct neuronal and brain networks to simulate the pathogenesis of the disease in its entirety and relevant connectivity, and to use the designed networks to computationally test for potential therapeutic strategies. Moreover, modeling the description of complex brain networks may also open new windows in our understanding of the basic mechanisms of the disease. In particular, solid and reliable computational models of the AD brain may help to elucidate the probabilities to which extent each of the hypotheses reviewed above contributes to the pathogenesis and progression of the disease.

In AD research computational approaches have been applied at various levels of complexity to investigate both single pathogenetic aspects of AD such as oligomerization, fibrillization, and aggregation of Aβ and AD-related proteins, Aβ binding motifs relevant to AD chemistry, structure – activity relationships and protein–protein interactions of AD relevant molecules, AD related cell signaling pathways, kinetics of enzyme substrate complexes relevant to APP processing, as well as to develop more complex systems such as molecular connectivity maps/protein interaction networks, neuronal and brain networks and their dynamics, gene–gene interactions and relevant proteomic changes, as well modeling of cognition/cognitive tasks.

In the following, a selection of recent advances made by applying computational approaches in AD-related research, will be presented; it is not intended to be complete or comprehensive.

Protein Aggregation

To understand the dynamics of the Aβ peptide aggregation process involved in AD [51.225] and the differentiation between disease and non-disease protein aggregation [51.226], molecular-level simulation models have been applied. Computationally derived structural models of Aβ and the interaction with possible aggregation inhibitors have been reported [51.227].

Protein–Protein Interactions

Examples of computational analysis and the prediction of protein–protein interaction include associations between the death cell receptor 6 (DR6) ectodomain and an N-terminal fragment of amyloid precursor protein (NAPP, [51.228]), in silico analysis of the apolipoprotein E and the Aβ peptide interaction [51.229], the effects of mutations on protein interactions [51.230], as well as the regulation of tau protein kinase II by its subunits [51.231].

AD-Related Cell Signaling Pathways

To identify protein signal pathways between APP and tau proteins, recently a modified network-constrained regularization analysis was applied to microarray data from a transgenic mouse model and AD patients. The corresponding protein pathways were then constructed by applying an integer linear programming model to integrate microarray data and the protein–protein interaction (PPI) database [51.232].

To quickly model and visualize alternative hypotheses about uncertain pathway knowledge, a prototype tool for representing and displaying cell-signaling pathway knowledge, for carrying out simple qualitative rea-

soning over these pathways and for generating quantitative biosimulation codes, *Chalkboard*, was developed and tested for the network of APP processing [51.233].

Structure – Activity Relationship of AD-Relevant Molecules

Using computational chemistry a multitarget quantitative structure-activity relationship (QSAR) model has recently been developed being able to predict the results of 42 different experimental tests for GSK-3 inhibitors with heterogeneous structural patterns [51.234], while computational studies of Cu(II)/Met and Cu(I)/Met binding motifs revealed a low affinity binding site of Cu(II) in Aβ [51.235].

Kinetics of Enzyme Substrate Complexes Relevant to APP Processing

Combining bioinformatics, molecular dynamics, and density functional theory studies, enabled researchers to elucidate the mechanisms for the hydrolytic cleavage of Val-Ile and Ala-Thr peptide bonds of APP by the intramembrane aspartyl protease presenilin 1 [51.236].

Gene–Gene Interactions

Uncovering genes that are linked with AD can help to understand pathogenesis and progression of the disease. In genome-wide association studies millions of single nucleotide polymorphisms (SNP) have been analyzed in thousands of AD patients and compared with healthy individuals in order to correlate genomic changes with the disease and to reveal harmful mutations or susceptibility genes [51.237]. As any single genetic variant for AD may be dependent on other genetic variants (gene–gene interaction) and environmental factors (gene-environment interaction) the evaluation of genetic association studies represents a computational challenge. This has been addressed by the introduction of the multifactor dimensionality reduction (MDR) method proposed by [51.238], which was further improved by the log-linear model-based multifactor dimensionality reduction (LM-MDR) method in sporadic AD [51.239], and grid computing to drive data intensive research [51.240]. Another computational strategy for simultaneously identifying multiple candidate genes for genetic human brain diseases from a brain-specific gene network-level perspective has been proposed by [51.241]. By integrating diverse genomic and proteomic datasets based on a Bayesian statistical model, a large-scale human brain-specific gene network has been established, which has been used to effectively identify multiple candidate genes for AD [51.241].

Gene–Protein Interactions, Posttranslational Modification of Gene Products

Microarray analyses are a valuable tool for large-scale screening of differential gene expressions that are presumably involved in AD pathogenesis, but cannot provide information about protein levels, posttranslational modifications of gene products, or activity changes, which are important for the understanding of the molecular processes of signal transduction pathways and intracellular interactions between proteins that influence cellular function. For example, toxicity studies of oligomeric Aβ (1–42) on cholinergic SN56.B5.G4 cells, revealed that gene products affected by Aβ (1–42) exposure were present in the endoplasmatic reticulum (ER), Golgi apparatus, and/or otherwise involved in protein modification and degradation (chaperones, ATF6) indicating a possible role of ER-mediated stress in Aβ-mediated toxicity. Moreover, a number of genes, which are known to be involved in AD (clusterin, acetylcholine transporter), were found to be affected following Aβ (1–42) exposure of cholinergic cells [51.242]. However, proteomic analyses of lysates of the cells exposed to Aβ (1–42) identified Aβ-mediated changes in protein levels and additionally in phosphorylation status, which provided complementary information to the microarray data. The levels of calreticulin and mitogen-activated protein kinase kinase 6c were upregulated in cholinergic cells exposed to Aβ (1–42), while γ-actin appeared downregulated. Decreased phosphorylation of phosphoproteins, such as the Rho GDP dissociation inhibitor, the ubiquitin carboxyl terminal hydrolase-1, and the tubulin α-chain isotype Mα6 have been observed [51.243]. The proteins identified have also been reported to be affected in brains of AD patients, suggesting a potential role of Aβ in influencing the integrity and functioning of the proteome in AD. The data suggest that Aβ (1–42) mediates its cytotoxic action by affecting proteins that also play key roles in a number of other physiological processes such as apoptosis, cytoskeletal changes, protein degradation of misfolded or damaged proteins, stress-related MAPK signaling, and GTPase function, indicating the Aβ-mediated disturbance of a complex mutually interacting network of signal transduction pathways. Pathological changes of one component of the network may have consequences on compartments of other cascades, finally resulting in either compensatory mechanisms, or when persistently overshooting, in pathogenic processes. In this respect, computational approaches may represent a very useful tool to model such complex networks because they are able to simulate the dynamics of

such networks and the consequences of any disturbance. Assessment of particular signaling cascades reveals kinetic parameters that can be entered into a more complex network composed of a number of interacting pathways. This means that the complexity of the system can be steadily increased, finally addressing not only static but modeling also dynamic features of the system.

Molecular Connectivity Maps/Protein Interaction Networks

A computational framework was recently developed to build disease-specific drug-protein connectivity maps by integration of gene/protein and drug connectivity information based on protein interaction networks and literature mining, without requiring information on gene expression profiles [51.244]. Such molecular connectivity maps of various diseases appear to be adequate tools to computationally prove the therapeutic profile of candidate drugs before application in clinical trials [51.244].

Neuronal and Brain Networks

There are different organizational levels by which brain networks can be designed comprising either single neurons (microscale), a group of neurons, or anatomically distinct brain regions (macroscale; for a review, see e.g., [51.245]). A brain network consists of two basic elements, the nodes representing neurons or regions of interest and edges linking the nodes. The edges define either functional or structural associations among different neuronal elements of the brain. The small-world network introduced by [51.246] has been found to be an attractive model for the characterization of complex brain networks because it captures both segregated and integrated information processing of the brain [51.247]. Based on EEG, MEG and/or fMRI data, and using modern graph theory-based computational approaches the designed brain network can be used to map brain functional activities under both normal and pathological conditions. Analysis of AD patients revealed altered small-world characteristics, which reflect the aberrant neuronal connectivity in AD brain and may explain the cognitive deficits [51.245].

Neural networks have also been used for longitudinal studies in AD [51.248]. The proposed new type of neural network (mixed effects neural network) has been found to be a reliable tool for predicting non-linear disease trajectories and uncovering significant prognostic factors in longitudinal databases of AD [51.248].

A computational model of network dynamics describing the progression of the neuropathology from the hippocampus into neocortical structures in AD has been proposed by [51.249]. The model focuses on a final common breakdown in function, termed runaway synaptic modification, and proposes that the imbalance of variables regulating the influence of synaptic transmission on synaptic modification causes the pathogenic processes in the hippocampus and cortex. Memory deficits are described as being due to increased interference effects on recent memory caused by runaway synaptic modification [51.250].

Applying the framework of an associative memory model, the interplay between synaptic deletion and compensation, and memory deterioration was computationally studied in AD [51.251]. The simulation shows that a disease-induced synaptic deletion may be compensated by a local, dynamic mechanism, where each neuron maintains the profile of its incoming postsynaptic current, suggesting that the primary factor in the pathogenesis of cognitive deficiencies in AD is the failure of local neuronal regulatory mechanisms [51.251].

Recently, a novel research framework for building probabilistic computational neurogenetic models (pCNGM) was proposed by [51.1]. The pCNGM represents a multilevel modeling framework inspired by the multilevel information processes in the brain. The framework comprises a set of several dynamic models including low (molecular) level models, a dynamic model of a protein regulatory network (PRN), and a probabilistic spiking neural network model (pSNN). It can be used for modeling both artificial cognitive systems and brain processes. Using data obtained from relevant genes/proteins a pCNGM may simulate their dynamic interaction which matches data related to brain development, higher-level brain function, or disorders such as AD [51.1].

Modeling of Cognition/Cognitive Tasks

There are a number of studies using computational modeling to simulate various cognitive and behavioral deficits observed in AD patients such as phonological dyslexia [51.252], decrease of performance in daily living activities [51.253], human path planning strategies [51.254], and memory impairments [51.255].

In conclusion, the recent advances in applying computational approaches to model the pathogenesis of AD by complex brain network methods provide some hope towards the understanding of the complexity of the disease as well as being able to computationally derive novel routes of treatment strategies and prove the therapeutic profile of candidate drugs.

51.9 General Conclusion: Mutual Interactions of Cholinergic, Aβ, and Vascular Pathologies

Taken together, the observations mentioned above indicate that a disbalance or disturbance of the homeostatic interrelationship between cholinergic function including NGF, APP metabolism, and control mechanisms of brain microvasculature (CBF, perfusion, glucose delivery) may result in deleterious consequences by favoring the amyloidogenic route of APP processing with enhanced plaque formation and induction of local neuroinflammatory events. Persisting β-amyloid-induced glial upregulation of pro-inflammatory cytokines then further contributes to damage cholinergic functions and further promotes the amyloidogenic pathway. On the other hand, aging and life-style associated damage of the brain microvasculature may affect Aβ clearance and perivascular drainage, promote cerebrovascular Aβ deposition, inducing partial loss of cholinergic vascular innervation and changes in vascular function, angiogenesis and VEGF upregulation with consequences on APP processing and Aβ accumulation, which finally may develop into a detrimental vicious cycle.

The interplay of the essential features observed in AD (Aβ accumulation, cholinergic dysfunction, vascu-

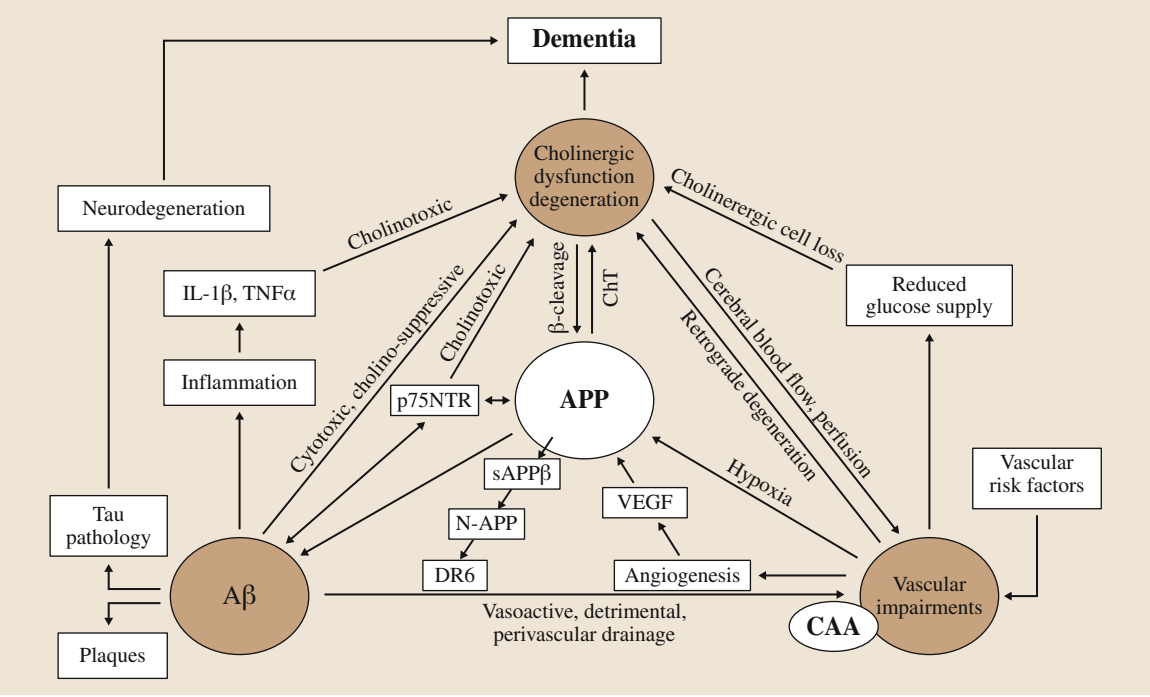

Fig. 51.6 *Interrelationship between cholinergic neurotransmission, APP metabolism, inflammation, and brain vascular system.* The interplay of the essential features observed in AD (Aβ accumulation, cholinergic dysfunction, vascular impairments, inflammation, neurodegeneration) and their mutually influencing processes is shown. A disbalance or disturbance of the homeostatic interrelationship between cholinergic function including NGF, APP metabolism, and control mechanisms of brain microvasculature (cerebral blood flow (CBF), perfusion, glucose delivery) may result in deleterious consequences by favoring the amyloidogenic route of APP processing with enhanced plaque formation and induction of local neuroinflammatory events. Persisting Aβ-induced glial upregulation of pro-inflammatory cytokines then further contributes to damage cholinergic functions as well as promotes the amyloidogenic pathway. On the other hand, aging, and life-style associated damage to the brain microvasculature may affect Aβ clearance and perivascular drainage, promote cerebrovascular Aβ deposition, inducing partial loss of cholinergic vascular innervation and changes in vascular function, angiogenesis, and VEGF upregulation with consequences on APP processing and Aβ accumulation, which finally may develop into a detrimental positive feedback loop, a vicious cycle

lar impairments, inflammation, neurodegeneration), and their mutually influencing processes are summarized in Fig. 51.6. We hypothesize that a disturbance of this homeostatic relationship may develop during late aging and may represent a major cause for the genesis of sporadic Alzheimer's disease. However, which kind of aging-related factors may play a role remains to be disclosed, but they would allow us to derive strategies for establishing prophylactic measures.

Computational approaches to model pathogenesis and progression of AD appear to be useful tools to reflect the disease in its entirety and internal connective relationships, and thus to open new views in our understanding of basic mechanisms of AD. The pCNGM research framework as recently introduced and described by [51.1], represents a promising and potential tool to model the AD brain in its complexity and dynamics at different operational levels, including functional neuron-glia-vascular units, gene–protein interactions, cell signaling cascades, regional association, and higher cognitive function. Thus computational approaches of modeling the AD brain may help to elucidate the probabilities to which extent each of the hypotheses reviewed in this chapter contribute to pathogenesis and progression of the disease.

References

51.1 N.K. Kasabov, R. Schliebs, H. Kojima: Probabilistic computational neurogenetic modeling: From cognitive systems to Alzheimer's disease, IEEE Trans. Auton. Mental Dev. **3**, 1–12 (2011)

51.2 A. Alzheimer: Über eine eigenartige Erkrankung der Hirnrinde, Allg. Z. Psychiatr. Psychisch-Gerichtl. Med. **64**, 146–148 (1907)

51.3 G.G. Glenner, C.W. Wong: Alzheimer's disease and Down's syndrome: Sharing of a unique cerebrovascular amyloid fibril protein, Biochem. Biophys. Res. Commun. **122**, 1131–1135 (1984)

51.4 C.L. Masters, G. Simms, N.A. Weinman, G. Multhaup, B.L. McDonald, K. Beyreuther: Amyloid plaque core protein in Alzheimer disease and down syndrome, Proc. Natl. Acad. Sci. USA **82**, 4245–4249 (1985)

51.5 J. Kang, H.G. Lemaire, A. Unterbeck, J.M. Salbaum, C.L. Masters, K.H. Grzeschik, G. Multhaup, K. Beyreuther, B. Müller-Hill: The precursor of Alzheimer's disease amyloid A4 protein resembles a cell-surface receptor, Nature **325**, 733–736 (1987)

51.6 D. Goldgaber, M.I. Lerman, O.W. McBride, U. Saffiotti, D.C. Gajdusek: (1987) Characterization and chromosomal localization of a cDNA encoding brain amyloid of Alzheimer's disease, Science **235**, 877–880 (2006)

51.7 N.K. Robakis, N. Ramakrishna, G. Wolfe, H.M. Wisniewski: Molecular cloning and characterization of a cDNA encoding the cerebrovascular and the neuritic plaque amyloid peptides, Proc. Natl. Acad. Sci. USA **84**, 4190–4194 (1987)

51.8 R.E. Tanzi, J.F. Gusella, P.C. Watkins, G.A. Bruns, P. St George-Hyslop, M.L. Van Keuren, D. Patterson, S. Pagan, D.M. Kurnit, R.L. Neve: Amyloid β protein gene: CDNA, mRNA distribution, and genetic linkage near the Alzheimer locus, Science **235**, 880–884 (1987)

51.9 H. Bickel: *Die Epidemiologie der Demenz*, Informationsblatt (Deutsche Alzheimer Gesellschaft e.V., Berlin 2010), available online at www.deutsche-alzheimer.de/

51.10 K. Herrup: Reimagining Alzheimer's disease–an age-based hypothesis, J. Neurosci. **30**, 16755–16762 (2010)

51.11 R.S. Turner: Alzheimer's disease, Semin. Neurol. **26**, 499–506 (2006)

51.12 R.J. Castellani, R.K. Rolston, M.A. Smith: Alzheimer disease, Dis. Mon. **56**, 484–546 (2010)

51.13 G.G. Glenner, C.W. Wong, V. Quaranta, E.D. Eanes: The amyloid deposits in Alzheimer's disease: Their nature and pathogenesis, Appl. Pathol. **2**, 357–369 (1984)

51.14 I. Grundke-Iqbal, K. Iqbal, Y.C. Tung, M. Quinlan, H.M. Wisniewski, L.I. Binder: Abnormal phosphorylation of the microtubule-associated protein tau (tau) in Alzheimer cytoskeletal pathology, Proc. Natl. Acad. Sci. USA **83**, 4913–4917 (1986)

51.15 I. Grundke-Iqbal, K. Iqbal, M. Quinlan, Y.C. Tung, M.S. Zaidi, H.M. Wisniewski: Microtubule-associated protein tau. A component of Alzheimer paired helical filaments, J. Biol. Chem. **261**, 6084–6089 (1986)

51.16 K. Iqbal, I. Grundke-Iqbal, A.J. Smith, L. George, Y.C. Tung, T. Zaidi: Identification and localization of a tau peptide to paired helical filaments of Alzheimer disease, Proc. Natl. Acad. Sci. USA **86**, 5646–5650 (1989)

51.17 R. Lee, P. Kermani, K.K. Teng, B.L. Hempstead: Regulation of cell survival by secreted proneurotrophins, Science **294**, 1945–1948 (2001)

51.18 K. Iqbal, C. Alonso Adel, S. Chen, M.O. Chohan, E. El-Akkad, C.X. Gong, S. Khatoon, B. Li, F. Liu, A. Rahman, H. Tanimukai, I. Grundke-Iqbal: Tau pathology in Alzheimer disease and other tauopathies, Biochim. Biophys. Acta **1739**, 198–210 (2005)

51.19 A. Alonso, T. Zaidi, M. Novak, I. Grundke-Iqbal, K. Iqbal: Hyperphosphorylation induces

self-assembly of tau into tangles of paired helical filaments/straight filaments, Proc. Natl. Acad. Sci. USA **98**, 6923–6928 (2001)

51.20 K.R. Patterson, C. Remmers, Y. Fu, S. Brooker, N.M. Kanaan, L. Vana, S. Ward, J.F. Reyes, K. Philibert, M.J. Glucksman, L.I. Binder: Characterization of prefibrillar tau oligomers in vitro and in Alzheimer disease, J. Biol. Chem. **286**, 23063–23076 (2011)

51.21 C. Ballard, S. Gauthier, A. Corbett, C. Brayne, D. Aarsland, E. Jones: Alzheimer's disease, Lancet **377**, 1019–1031 (2011)

51.22 M.S. Wolfe: Tau mutations in neurodegenerative diseases, J. Biol. Chem. **284**, 6021–6025 (2009)

51.23 M. Goedert, M.G. Spillantini: A century of Alzheimer's disease, Science **314**, 777–781 (2006)

51.24 S.A. Small, K. Duff: Linking Aβ and tau in late-onset Alzheimer's disease: A dual pathway hypothesis, Neuron **60**, 534–542 (2008)

51.25 L. Bertram, C.M. Lill, R.E. Tanzi: The genetics of Alzheimer disease: Back to the future, Neuron **68**, 270–281 (2010)

51.26 E.H. Corder, A.M. Saunders, W.J. Strittmatter, D.E. Schmechel, P.C. Gaskell, G.W. Small, A.D. Roses, J.L. Haines, M.A. Pericak-Vance: Gene dose of apolipoprotein E type 4 allele and the risk of Alzheimer's disease in late onset families, Science **261**, 921–923 (1993)

51.27 A.M. Saunders, W.J. Strittmatter, D. Schmechel, P.H. George-Hyslop, M.A. Pericak-Vance, S.H. Joo, B.L. Rosi, J.F. Gusella, D.R. Crapper-MacLachlan, M.J. Alberts, C. Hulette, B. Crain, D. Goldgaber, A.D. Roses: Association of apolipoprotein E allele epsilon 4 with late-onset familial and sporadic Alzheimer's disease, Neurology **43**, 467–1472 (1993)

51.28 Y. Huang: Aβ-independent roles of apolipoprotein E4 in the pathogenesis of Alzheimer's disease, Trends Mol. Med. **16**, 287–294 (2010)

51.29 E. Rogaeva, Y. Meng, J.H. Lee, Y. Gu, T. Kawarai, F. Zou, T. Katayama, C.T. Baldwin, R. Cheng, H. Hasegawa, F. Chen, N. Shibata, K.L. Lunetta, R. Pardossi-Piquard, C. Bohm, Y. Wakutani, L.A. Cupples, K.T. Cuenco, R.C. Green, L. Pinessi, I. Rainero, S. Sorbi, A. Bruni, R. Duara, R.P. Friedland, R. Inzelberg, W. Hampe, H. Bujo, Y.Q. Song, O.M. Andersen, T.E. Willnow, N. Graff-Radford, R.C. Petersen, D. Dickson, S.D. Der, P.E. Fraser, G. Schmitt-Ulms, S. Younkin, R. Mayeux, L.A. Farrer, P. St George-Hyslop: The neuronal sortilin-related receptor SORL1 is genetically associated with Alzheimer disease, Nat. Genet. **39**, 168–177 (2007)

51.30 K. Morgan: The three new pathways leading to AD, Neuropathol. Appl. Neurobiol. **37**, 353–357 (2011)

51.31 D.M. Bowen, C.B. Smith, P. White, A.N. Davison: Neurotransmitter-related enzymes and indices of hypoxia in senile dementia and other abiotrophies, Brain **99**, 459–496 (1976)

51.32 P. Davies, A.J. Maloney: Selective loss of central cholinergic neurons in Alzheimer's disease, Lancet **2**, 1403 (1976)

51.33 E.K. Perry, P.H. Gibson, G. Blessed, R.H. Perry, B.E. Tomlinson: Neurotransmitter enzyme abnormalities in senile dementia. Choline acetyltransferase and glutamic acid decarboxylase activities in necropsy brain tissue, J. Neurol. Sci. **34**, 247–265 (1977)

51.34 E.K. Perry, R.H. Perry, G. Blessed, B.E. Tomlinson: Necropsy evidence of central cholinergic deficits in senile dementia, Lancet. **1**, 189 (1977)

51.35 D.S. Auld, T.J. Kornecook, S. Bastianetto, R. Quirion: Alzheimer's disease and the basal forebrain cholinergic system: Relations to β-amyloid peptides, cognition, and treatment strategies, Prog. Neurobiol. **68**, 209–245 (2002)

51.36 A. Nordberg: Neuroreceptor changes in Alzheimer disease, Cerebrovasc. Brain Metab. Rev. **4**, 303–328 (1992)

51.37 L.M. Bierer, V. Haroutunian, S. Gabriel, P.J. Knott, L.S. Carlin, D.P. Purohit, D.P. Perl, J. Schmeidler, P. Kanof, K.L. Davis: Neurochemical correlates of dementia severity in Alzheimer's disease: Relative importance of the cholinergic deficits, J. Neurochem. **64**, 749–760 (1995)

51.38 W. Gsell, G. Jungkunz, P. Riederer: Functional neurochemistry of Alzheimer's disease, Curr. Pharmacol. Des. **10**, 265–293 (2004)

51.39 R.T. Bartus: On neurodegenerative diseases, models, and treatment strategies: Lessons learned and lessons forgotten a generation following the cholinergic hypothesis, Exp. Neurol. **163**, 495–529 (2000)

51.40 E.J. Mufson, S.E. Counts, S.E. Perez, S.D. Ginsberg: Cholinergic system during the progression of Alzheimer's disease: Therapeutic implications, Exp, Rev. Neurother. **8**, 1703–1718 (2008)

51.41 E.J. Mufson, S.E. Counts, M. Fahnestock, S.D. Ginsberg: Cholinotrophic molecular substrates of mild cognitive impairment in the elderly, Curr. Alzheimer Res. **4**, 340–350 (2007)

51.42 A.C. Cuello, M.A. Bruno, K.F. Bell: NGF-cholinergic dependency in brain aging, MCI and Alzheimer's disease, Curr. Alzheimer Res. **4**, 351–358 (2007)

51.43 Y.W. Zhang, R. Thompson, H. Zhang, H. Xu: APP processing in Alzheimer's disease, Mol. Brain **4**, 3 (2011), doi: 10.1186/1756-6606-4-3

51.44 A. Nikolaev, T. McLaughlin, D.D. O'Leary, M. Tessier-Lavigne: APP binds DR6 to trigger axon pruning and neuron death via distinct caspases, Nature **457**, 981–989 (2009)

51.45 H. Li, B. Wang, Z. Wang, Q. Guo, K. Tabuchi, R.E. Hammer, T.C. Südhof, H. Zheng: Soluble amyloid precursor protein (APP) regulates transthyretin and Klotho gene expression without rescuing the essential function of APP, Proc. Natl. Acad. Sci. USA **107**, 17362–17367 (2010)

51.46 B. De Strooper, R. Vassar, T. Golde: The secretases: Enzymes with therapeutic potential in Alzheimer disease, Nat. Rev. Neurol. **6**, 99–107 (2010)

51.47 J. Hardy, D. Allsop: Amyloid deposition as the central event in the aetiology of Alzheimer's disease, Trends Pharmacol. Sci. **12**, 383–388 (1991)

51.48 J. Hardy: The amyloid hypothesis for Alzheimer's disease: A critical reappraisal, J. Neurochem. **110**, 1129–1134 (2009)

51.49 B.K. Harvey, C.T. Richie, B.J. Hoffer, M. Airavaara: Transgenic animal models of neurodegeneration based on human genetic studies, J. Neural Transm. **118**, 27–45 (2011)

51.50 A. Thathiah, B. De Strooper: The role of G protein-coupled receptors in the pathology of Alzheimer's disease, Nat. Rev. Neurosci. **12**, 73–87 (2011)

51.51 M. Pákáski, J. Kálmán: Interactions between the amyloid and cholinergic mechanisms in Alzheimer's disease, Neurochem. Int. **53**, 103–111 (2008)

51.52 R.H. Parri, T.K. Dineley: Nicotinic acetylcholine receptor interaction with beta-amyloid: Molecular, cellular, and physiological consequences, Curr. Alzheimer Res. **7**, 27–39 (2010)

51.53 M.M. Mesulam: Alzheimer plaques and cortical cholinergic innervation, Neuroscience **17**, 275–276 (1986)

51.54 M.A. Moran, E.J. Mufson, P. Gomez-Ramos: Colocalization of cholinesterases with β-amyloid protein in aged and Alzheimer's brain, Acta Neuropathol. **85**, 362–369 (1993)

51.55 R.M. Nitsch, B.E. Slack, R.J. Wurtman, J.H. Growdon: Release of Alzheimer amyloid precursor derivatives stimulated by activation of muscarinic acetylcholine receptors, Science **258**, 304–307 (1992)

51.56 J.D. Buxbaum, M. Oishi, H.I. Chen, R. Pinkas-Kramarski, E.A. Jaffe, S.E. Gandy, P. Greengard: Cholinergic agonists and interleukin 1 regulate processing and secretion of the Alzheimer β/A4 amyloid protein precursor, Proc. Natl. Acad. Sci. USA **89**, 10075–10078 (1992)

51.57 R.M. Nitsch, S.A. Farber, J.H. Growdon, R.J. Wurtman: Release of amyloid beta-protein precursor derivatives by electrical depolarization of rat hippocampal slices, Proc. Natl. Acad. Sci. USA **90**, 5191–5193 (1993)

51.58 H.A. Ensinger, H.N. Doods, A.R. Immel-Sehr, F.J. Kuhn, G. Lambrecht, K.D. Mendla, R.E. Muller, E. Mutschler, A. Sagrada, G. Walther: WAL 2014–a muscarinic agonist with preferential neuron-stimulating properties, Life Sci. **52**, 473–480 (1993)

51.59 S.A. Farber, R.M. Nitsch, J.G. Schulz, R.J. Wurtman: Regulated secretion of beta-amyloid precursor protein in rat brain, J. Neurosci. **15**, 7442–7451 (1995)

51.60 A. Walland, S. Burkard, R. Hammer, W. Troger: In vivo consequences of M1-receptor activation by talsaclidine, Life Sci. **60**, 977–984 (1997)

51.61 D.M. Müller, K. Mendla, S.A. Farber, R.M. Nitsch: Muscarinic M1 receptor agonists increase the secretion of the amyloid precursor protein ectodomain, Life Sci. **60**, 985–991 (1997)

51.62 C. Hock, A. Maddalena, A. Raschig, F. Müller-Spahn, G. Eschweiler, K. Hager, I. Heuser, H. Hampel, T. Müller-Thomsen, W. Oertel, M. Wienrich, A. Signorell, C. Gonzalez-Agosti, R.M. Nitsch: Treatment with the selective muscarinic m1 agonist talsaclidine decreases cerebrospinal fluid levels of A beta 42 in patients with Alzheimer's disease, Amyloid **10**, 1–6 (2003)

51.63 R.M. Nitsch: Treatment with the selective muscarinic m1 agonist talsaclidine decreases cerebrospinal fluid levels of Aβ42 in patients with Alzheimer's disease, Amyloid **10**, 1–6 (2003)

51.64 A.Y. Hung, C. Haass, R.M. Nitsch, W.Q. Qiu, M. Citron, R.J. Wurtman, J.H. Growdon, D.J. Selkoe: Activation of protein kinase C inhibits cellular production of the amyloid beta-protein, J. Biol. Chem. **268**, 22959–22962 (1993)

51.65 M. Cisse, U. Braun, M. Leitges, A. Fisher, G. Pages, F. Checler, B. Vincent: ERK1-independent α-secretase cut of β-amyloid precursor protein via M1 muscarinic receptors and PKCα/ε, Mol. Cell Neurosci. **47**, 223–232 (2011)

51.66 B.E. Slack, J. Breu, M.A. Petryniak, K. Srivastava, R.J. Wurtman: Tyrosine phosphorylation-dependent stimulation of amyloid precursor protein secretion by the m3 muscarinic acetylcholine receptor, J. Biol. Chem. **270**, 8337–8344 (1995)

51.67 R. Haring, A. Fisher, D. Marciano, Z. Pittel, Y. Kloog, A. Zuckerman, N. Eshhar, E. Heldman: Mitogen-activated protein kinase-dependent and protein kinase C-dependent pathways link the m1 muscarinic receptor to beta-amyloid precursor protein secretion, J. Neurochem. **71**, 2094–2103 (1998)

51.68 S. Roßner, U. Ueberham, R. Schliebs, J.R. Perez-Polo, V. Bigl: The regulation of amyloid precursor protein metabolism by cholinergic mechanisms and neurotrophin receptor signaling, Prog. Neurobiol. **56**, 541–569 (1998)

51.69 L. Lin, B. Georgievska, A. Mattsson, O. Isacson: Cognitive changes and modified processing of amyloid precursor protein in the cortical and hippocampal system after cholinergic synapse loss and muscarinic receptor activation, Proc. Natl. Acad. Sci. USA **96**, 12108–12113 (1999)

51.70 H. Seo, A.W. Ferree, O. Isacson: Cortico-hippocampal APP and NGF levels are dynamically altered by cholinergic muscarinic antagonist or M1 agonist treatment in normal mice, Eur J. Neurosci. **15**, 498–505 (2002)

51.71 W. Liskowsky, R. Schliebs: Muscarinic acetylcholine receptor inhibition in transgenic Alzheimer-like Tg2576 mice by scopolamine favours the amyloidogenic route of processing of amyloid precursor protein, Int. J. Devl. Neurosci. **24**, 149–156 (2006)

51.72 A.A. Davis, J.J. Fritz, J. Wess, J.J. Lah, A.I. Levey: Deletion of M1 muscarinic acetylcholine receptors increases amyloid pathology in vitro and in vivo, J. Neurosci. **30**, 4190–4196 (2010)

51.73 A. Caccamo, S. Oddo, L.M. Billings, K.N. Green, H. Martinez-Coria, A. Fisher, F.M. LaFerla: M1 receptors play a central role in modulating AD-like pathology in transgenic mice, Neuron **49**, 671–682 (2006)

51.74 R. Medeiros, M. Kitazawa, A. Caccamo, D. Baglietto-Vargas, T. Estrada-Hernandez, D.H. Cribbs, A. Fisher, F.M. Laferla: Loss of muscarinic M(1) receptor exacerbates Alzheimer's disease-like pathology and cognitive decline, Am. J. Pathol. **179**, 980–991 (2011)

51.75 T. Züchner, J.R. Perez-Polo, R. Schliebs: β-secretase BACE1 is differentially controlled through muscarinic acetylcholine receptor signaling, J. Neurosci. Res. **77**, 250–257 (2004)

51.76 T.A. Kohout, R.J. Lefkowitz: Regulation of G protein-coupled receptor kinases and arrestins during receptor desensitization, Mol. Pharmacol. **63**, 9–18 (2003)

51.77 J. Liu, I. Rasul, Y. Sun, G. Wu, L. Li, R.T. Premont, W.Z. Suo: GRK5 deficiency leads to reduced hippocampal acetylcholine level via impaired presynaptic M2/M4 autoreceptor desensitization, J. Biol. Chem. **284**, 19564–19571 (2009)

51.78 W. Zhang, A.S. Basile, J. Gomeza, L.A. Volpicelli, A.I. Levey, J. Wess: Characterization of central inhibitory muscarinic autoreceptors by the use of muscarinic acetylcholine receptor knock-out mice, J. Neurosci. **22**, 1709–1717 (2002)

51.79 Z. Suo, M. Wu, B.A. Citron, G.T. Wong, B.W. Festoff: Abnormality of G-protein-coupled receptor kinases at prodromal and early stages of Alzheimer's disease: An association with early beta-amyloid accumulation, J. Neurosci. **24**, 3444–3452 (2004)

51.80 S. Cheng, L. Li, S. He, J. Liu, Y. Sun, M. He, K. Grasing, R.T. Premont, W.Z. Suo: GRK5 deficiency accelerates β-amyloid accumulation in Tg2576 mice via impaired cholinergic activity, J. Biol. Chem. **285**, 41541–41548 (2010)

51.81 S. Efthimiopoulos, D. Vassilacopoulou, J.A. Ripellino, N. Tezapsidis, N.K. Robakis: Cholinergic agonists stimulate secretion of soluble full-length amyloid precursor protein in neuroendocrine cells, Proc. Natl. Acad. Sci. USA **93**, 8046–8050 (1996)

51.82 S.H. Kim, Y.K. Kim, S.J. Jeong, C. Haass, Y.H. Kim, Y.H. Suh: Enhanced release of secreted form of Alzheimer's amyloid precursor protein from PC12 cells by nicotine, Mol. Pharmacol. **52**, 430–436 (1997)

51.83 J. Seo, S. Kim, H. Kim, C.H. Park, S. Jeong, J. Lee, S.H. Choi, K. Chang, J. Rah, J. Koo, E. Kim, Y. Suh: Effects of nicotine on APP secretion and Aβ- or CT(105)-induced toxicity, Biol. Psychiatry **49**, 240–247 (2001)

51.84 X.L. Qi, A. Nordberg, J. Xiu, Z.Z. Guan: The consequences of reducing expression of the alpha7 nicotinic receptor by RNA interference and of stimulating its activity with an alpha7 agonist in SH-SY5Y cells indicate that this receptor plays a neuroprotective role in connection with the pathogenesis of Alzheimer's disease, Neurochem. Int. **51**, 377–383 (2007)

51.85 T. Utsuki, M. Shoaib, H.W. Holloway, D.K. Ingram, W.C. Wallace, V. Haroutunian, K. Sambamurti, D.K. Lahiri, N.H. Greig: Nicotine lowers the secretion of the Alzheimer's amyloid β-protein precursor that contains amyloid β-peptide in rat, J. Alzheimers Dis. **4**, 405–415 (2002)

51.86 D.K. Lahiri, T. Utsuki, D. Chen, M.R. Farlow, M. Shoaib, D.K. Ingram, N.H. Greig: Nicotine reduces the secretion of Alzheimer's β-amyloid precursor protein containing β-amyloid peptide in the rat without altering synaptic proteins, Ann. N. Y. Acad. Sci. **965**, 364–372 (2002)

51.87 M. Mousavi, E. Hellström-Lindahl: Nicotinic receptor agonists and antagonists increase sAPPalpha secretion and decrease Abeta levels in vitro, Neurochem. Int. **54**, 237–244 (2009)

51.88 H.Z. Nie, S. Shi, R.J. Lukas, W.J. Zhao, Y.N. Sun, M. Yin: Activation of α7 nicotinic receptor affects APP processing by regulating secretase activity in SH-EP1-α7 nAChR-hAPP695 cells, Brain Res. **1356**, 112–120 (2010)

51.89 H.Z. Nie, Z.Q. Li, Q.X. Yan, Z.J. Wang, W.J. Zhao, L.C. Guo, M. Yin: Nicotine decreases beta-amyloid through regulating BACE1 transcription in SH-EP1-$\alpha4\beta2$ nAChR-APP695 cells, Neurochem. Res. **36**, 904–912 (2011)

51.90 E. Hellstrom-Lindahl, J. Court, J. Keverne, M. Svedberg, M. Lee, A. Marutle, A. Thomas, E. Perry, I. Bednar, A. Nordberg: Nicotine reduces Aβ in the brain and cerebral vessels of APPsw mice, Eur. J. Neurosci. **19**, 2703–2710 (2004)

51.91 M. Srivareerat, T.T. Tran, S. Salim, A.M. Aleisa, K.A. Alkadhi: Chronic nicotine restores normal Aβ levels and prevents short-term memory and E-LTP impairment in Aβ rat model of Alzheimer's disease, Neurobiol. Aging **32**, 834–844 (2011)

51.92 J.A. Court, M. Johnson, D. Religa, J. Keverne, R. Kalaria, E. Jaros, I.G. McKeith, R. Perry, J. Naslund, E.K. Perry: Attenuation of Aβ deposition in the entorhinal cortex of normal elderly individuals associated with tobacco smoking, Neuropathol. Appl. Neurobiol. **31**, 522–535 (2005)

51.93 A.M. Klein, N.W. Kowall, R.J. Ferrante: Neurotoxicity and oxidative damage of β-amyloid 1–42 versus β-amyloid 1–40 in the mouse cerebral cortex, Ann. N. Y. Acad. Sci. **893**, 314–320 (1999)

51.94 W. Härtig, A. Bauer, K. Brauer, J. Gosche, T. Hortobagyi, B. Penke, R. Schliebs, T. Harkany: Functional recovery of cholinergic basal forebrain neurons

under disease conditions: Old problems, Rev. Neurosci. **13**, 95–165 (2002)
51.95 D.M. Walsh, D.J. Selkoe: Aβ oligomers – a decade of discovery, J. Neurochem. **101**, 1172–1184 (2007)
51.96 B.A. Yankner, T. Lu: Amyloid β-protein toxicity and the pathogenesis of Alzheimer disease, J. Biol. Chem. **284**, 4755–4759 (2009)
51.97 R. Schliebs, T. Arendt: The cholinergic system in aging and neuronal degeneration, Behav. Brain Res. **221**, 555–563 (2011)
51.98 T. Ondrejcak, I. Klyubin, N.W. Hu, A.E. Barry, W.K. Cullen, M.J. Rowan: Alzheimer's disease amyloid beta-protein and synaptic function, Neuromolecular Med. **12**, 13–26 (2010)
51.99 M.S. Parihar, G.J. Brewer: Amyloid beta as a modulator of synaptic plasticity, J. Alzheimers Dis. **22**, 741–763 (2010)
51.100 J.P. Cleary, D.M. Walsh, J.J. Hofmeister, G.M. Shankar, M.A. Kuskowski, D.J. Selkoe, K.H. Ashe: Natural oligomers of the amyloid-β protein specifically disrupt cognitive function, Nat. Neurosci. **8**, 79–84 (2005)
51.101 S. Lesné, M.T. Koh, L. Kotinilek, R. Kayed, C.G. Glabe, A. Yang, M. Gallagher, K.H. Ashe: A specific amyloid protein assembly in the brain impairs memory, Nature **440**, 352–357 (2006)
51.102 C.A. McLean, R.A. Cherny, F.W. Fraser, S.J. Fuller, M.J. Smith, K. Beyreuther, A.I. Bush, C.L. Masters: Soluble pool of Aβ amyloid as a determinant of severity of neurodegeneration in Alzheimer's disease, Ann. Neurol. **46**, 860–866 (1999)
51.103 J. Apelt, A. Kumar, R. Schliebs: Impairment of cholinergic neurotransmission in adult and aged transgenic Tg2576 mouse brain expressing the Swedish mutation of human beta-amyloid precursor protein, Brain Res. **953**, 17–30 (2002)
51.104 H.J. Lüth, J. Apelt, A. Ihunwo, R. Schliebs: Degeneration of β-amyloid-associated cholinergic structures in transgenic APPSW mice, Brain Res. **977**, 16–22 (2003)
51.105 M. Klingner, J. Apelt, A. Kumar, D. Sorger, O. Sabri, J. Steinbach, M. Scheunemann, R. Schliebs: Alterations in cholinergic and non-cholinergic neurotransmitter receptor densities in transgenic Tg2576 mouse brain with β-amyloid plaque pathology, Int. J. Devl. Neurosci. **21**, 357–369 (2003)
51.106 A. Bellucci, I. Luccarini, C. Scali, C. Prosperi, M.G. Giovannini, G. Pepeu, F. Casamenti: Cholinergic dysfunction, neuronal damage and axonal loss in TgCRND8 mice, Neurobiol. Dis. **23**, 260–272 (2006)
51.107 D. Van Dam, B. Marescau, S. Engelborghs, T. Cremers, J. Mulder, M. Staufenbiel, P.P. De Deyn: Analysis of cholinergic markers, biogenic amines, and amino acids in the CNS of two APP overexpression mouse models, Neurochem. Int. **46**, 409–422 (2005)
51.108 K.R. Bales, E.T. Tzavara, S. Wu, M.R. Wade, F.P. Bymaster, S.M. Paul, G.G. Nomikos: Cholinergic dysfunction in a mouse model of Alzheimer disease is reversed by an anti-Aβ antibody, J. Clin. Invest. **116**, 825–832 (2006)
51.109 E. Machová, J. Jakubík, P. Michal, M. Oksman, H. Iivonen, H. Tanila, V. Dolezal: Impairment of muscarinic transmission in transgenic APPswe/PS1dE9 mice, Neurobiol. Aging. **29**, 368–378 (2008)
51.110 Y. Luo, B. Bolon, S. Kahn, B.D. Bennett, S. Babu-Khan, P. Denis, W. Fan, H. Kha, J. Zhang, Y. Gong, L. Martin, J.C. Louis, Q. Yan, W.G. Richards, M. Citron, R. Vassar: Mice deficient in BACE1, the Alzheimer's beta-secretase, have normal phenotype and abolished β-amyloid generation, Nat. Neurosci. **4**, 231–232 (2001)
51.111 M. Ohno, E.A. Sametsky, L.H. Younkin, H. Oakley, S.G. Younkin, M. Citron, R. Vassar, J.F. Disterhoft: BACE1 deficiency rescues memory deficits and cholinergic dysfunction in a mouse model of Alzheimer's disease, Neuron. **41**, 27–33 (2004)
51.112 F. Bao, L. Wicklund, P.N. Lacor, W.L. Klein, A. Nordberg, A. Marutle: Different β-amyloid oligomer assemblies in Alzheimer brains correlate with age of disease onset and impaired cholinergic activity, Neurobiol. Aging. **33**, 825.e1–825.e13 (2012)
51.113 C. Haass, D.J. Selkoe: Cellular processing of beta-amyloid precursor protein and the genesis of amyloid beta-peptide, Cell **75**, 1039–1042 (1993)
51.114 M. Shoji, T.E. Golde, J. Ghiso, T.T. Cheung, S. Estus, L.M. Shaffer, X.D. Cai, D.M. McKay, R. Tintner, B. Frangione: Production of the Alzheimer amyloid beta protein by normal proteolytic processing, Science **258**, 126–129 (1992)
51.115 S. Kar, S.P. Slowikowski, D. Westaway, H.T. Mount: Interactions between β-amyloid and central cholinergic neurons: Implications for Alzheimer's disease, J. Psychiatry Neurosci. **29**, 427–441 (2004)
51.116 R.J. Wurtman: Choline metabolism as a basis for the selective vulnerability of cholinergic neurons, Trends Neurosci. **15**, 117–122 (1992)
51.117 C. Hock, A. Maddalena, A. Raschig, F. Muller-Spahn, G. Eschweiler, K. Hager, I. Heuser, H. Hampel, T. Muller-Thomsen, W. Oertel, M. Wienrich, A. Signorell, C. Gonzalez-Agosti, M. Hoshi, A. Takashima, M. Murayama, K. Yasutake, N. Yoshida, K. Ishiguro, T. Hoshino, K. Imahori: Nontoxic amyloid β peptide 1–42 suppresses acetylcholine synthesis. Possible role in cholinergic dysfunction in Alzheimer's disease, J. Biol. Chem. **272**, 2038–2041 (1997)
51.118 W.A. Pedersen, M.A. Kloczewiak, J.K. Blusztajn: Amyloid β-protein reduces acetylcholine synthesis in a cell line derived from cholinergic neurons of the basal forebrain, Proc. Natl. Acad. Sci. USA **93**, 8068–8071 (1996)
51.119 D. Hicks, D. John, N.Z. Makova, Z. Henderson, N.N. Nalivaeva, A.J. Turner: Membrane targeting, J. Neurochem. **116**, 742–746 (2011)

51.120 J.F. Kelly, K. Furukawa, S.W. Barger, M.R. Rengen, R.J. Mark, E.M. Blanc, G.S. Roth, M.P. Mattson: Amyloid β-peptide disrupts carbachol-induced muscarinic cholinergic signal transduction in cortical neurons, Proc. Natl. Acad. Sci. USA **93**, 6753–6758 (1996)

51.121 B. Wang, L. Yang, Z. Wang, H. Zheng: Amyolid precursor protein mediates presynaptic localization and activity of the high-affinity choline transporter, Proc. Natl. Acad. Sci. USA **104**, 14140–14145 (2007)

51.122 J. Pavia, J. Alberch, I. Alvárez, A. Toledano, M.L. de Ceballos: Repeated intracerebroventricular administration of beta-amyloid (25–35) to rats decreases muscarinic receptors in cerebral cortex, Neurosci. Lett. **278**, 69–72 (2000)

51.123 A. Thathiah, B. De Strooper: G protein-coupled receptors, cholinergic dysfunction, and Aβ toxicity in Alzheimer's disease, Sci. Signal. **2**(93), re8 (2009)

51.124 T. Chiba, M. Yamada, J. Sasabe, K. Terashita, M. Shimoda, M. Matsuoka, S. Aiso: Amyloid-β causes memory impairment by disturbing the JAK2/STAT3 axis in hippocampal neurons, Mol. Psychiatry **14**, 206–222 (2009)

51.125 L. Burghaus, U. Schütz, U. Krempel, R.A. de Vos, E.N. Jansen Steur, A. Wevers, J. Lindstrom, H. Schröder: Quantitative assessment of nicotinic acetylcholine receptor proteins in the cerebral cortex of Alzheimer patients, Mol. Brain Res. **76**, 385–388 (2000)

51.126 M. Mousavi, E. Hellström-Lindahl, Z.Z. Guan, K.R. Shan, R. Ravid, A. Nordberg: Protein and mRNA levels of nicotinic receptors in brain of tobacco using controls and patients with Alzheimer's disease, Neuroscience **122**, 515–520 (2003)

51.127 C.M. Martin-Ruiz, J.A. Court, E. Molnar, M. Lee, C. Gotti, A. Mamalaki, T. Tsouloufis, S. Tzartos, C. Ballard, R.H. Perry, E.K. Perry: Alpha4 but not alpha3 and alpha7 nicotinic acetylcholine receptor subunits are lost from the temporal cortex in Alzheimer's disease, J. Neurochem. **73**, 1635–1640 (1999)

51.128 A. Wevers, H. Schröder: Nicotinic acetylcholine receptors in Alzheimer's disease, J. Alzheimers Dis. **1**, 207–219 (1999)

51.129 A. Nordberg: Nicotinic receptor abnormalities of Alzheimer's disease: Therapeutic implications, Biol. Psychiatry **49**, 200–210 (2001)

51.130 S.D. Buckingham, A.K. Jones, L.A. Brown, D.B. Sattelle: Nicotinic acetylcholine receptor signalling: Roles in Alzheimer's disease and amyloid neuroprotection, Pharmacol. Rev. **61**, 39–61 (2009)

51.131 S. Jürgensen, S. Ferreira: Nicotinic receptors, amaloid-β, and synaptic failure in Alzheimer's disease, J. Mol. Neurosci. **40**, 221–229 (2010)

51.132 R.G. Nagele, M.R. D'Andrea, W.J. Anderson, H.Y. Wang: Intracellular accumulation of beta-amyloid (1–42) in neurons is facilitated by the alpha 7 nicotinic acetylcholine receptor in Alzheimer's disease, Neuroscience **110**, 199–211 (2002)

51.133 F.J. Barrantes, V. Borroni, S. Vallés: Neuronal nicotinic acetylcholine receptor-cholesterol crosstalk in Alzheimer's disease, FEBS Lett. **584**, 1856–1863 (2010)

51.134 Z.Z. Guan, H. Miao, J.Y. Tian, C. Unger, A. Nordberg, X. Zhang: Suppressed expression of nicotinic acetylcholine receptors by nanomolar β-amyloid peptides in PC12 cells, J. Neural Transm. **108**, 1417–1433 (2001)

51.135 Q. Liu, H. Kawai, D.K. Berg: β-amyloid peptide blocks the response of α7-containing nicotinic receptors on hippocampal neurons, Proc. Natl. Acad. Sci. USA **98**, 4734–4739 (2001)

51.136 K.A. Alkadhi, K.H. Alzoubi, M. Srivareerat, T.T. Tran: Elevation of BACE in an Aβ rat model of Alzheimer's disease: Exacerbation by chronic stress and prevention by nicotine, Int. J. Neuropsychopharmacol. **28**, 1–11 (2011)

51.137 H.Y. Wang, D.H. Lee, M.R. D'Andrea, P.A. Peterson, R.P. Shank, A.B. Reitz: β-Amyloid(1–42) binds to α7 nicotinic acetylcholine receptor with high affinity, Implications for Alzheimer's disease pathology, J. Biol. Chem. **275**, 5626–5632 (2000)

51.138 H.Y. Wang, D.H. Lee, C.B. Davis, R.P. Shank: Amyloid peptide Aβ (1–42) binds selectively and with picomolar affinity to α7 nicotinic acetylcholine receptors, J. Neurochem. **75**, 1155–1161 (2000)

51.139 S. Oddo, A. Caccamo, K.N. Green, K. Liang, L. Tran, Y. Chen, F.M. Leslie, F.M. LaFerla: Chronic nicotine administration exacerbates tau pathology in a transgenic model of Alzheimer's disease, Proc. Natl. Acad. Sci. USA **102**, 3046–3051 (2005)

51.140 X.L. Qi, J. Xiu, K.R. Shan, Y. Xiao, R. Gu, R.Y. Liu, Z.Z. Guan: Oxidative stress induced by β-amyloid peptide (1–42) is involved in the altered composition of cellular membrane lipids and the decreased expression of nicotinic receptors in human SH-SY5Y neuroblastoma cells, Neurochem. Int. **46**, 613–621 (2005)

51.141 Z.Z. Guan, W.F. Yu, K.R. Shan, T. Nordman, J. Olsson, A. Nordberg: Loss of nicotinic receptors induced by β-amyloid peptides in PC12 cells: Possible mechanism involving lipid peroxidation, J. Neurosci. Res. **71**, 397–406 (2003)

51.142 K.T. Dineley, M. Westerman, D. Bui, K. Bell, K.H. Ashe, J.D. Sweatt: β-amyloid activates the mitogen-activated protein kinase cascade via hippocampal α7 nicotinic acetylcholine receptors: In vitro and in vivo mechanisms related to Alzheimer's disease, J. Neurosci. **21**, 4125–4133 (2001)

51.143 H.Y. Wang, W. Li, N.J. Benedetti, D.H. Lee: α7 nicotinic acetylcholine receptors mediate β-amyloid peptide-induced tau protein phosphorylation, J. Biol. Chem. **278**, 31547–31553 (2003)

51.144 K.T. Dineley, K.A. Bell, D. Bui, J.D. Sweatt: β-Amyloid peptide activates α7 nicotinic acetylcholine receptors expressed in *Xenopus oocytes*, J. Biol. Chem. **277**, 25056–25061 (2002)

51.145 M. Bencherif, P. Lippiello: α7 neuronal nicotinic receptors: The missing link to understanding Alzheimer's etiopathology?, Med. Hypotheses **74**, 281–285 (2010)

51.146 M. Hu, J.F. Waring, M. Gopalakrishnan, J. Li: Role of GSK-3β activation and α7 nAChRs in $A\beta_{1-42}$-induced tau phosphorylation in PC12 cells, J. Neurochem. **106**, 1371–1377 (2008)

51.147 G. Dziewczapolski, C.M. Glogowski, E. Masliah, S.F. Heinemann: Deletion of the α7 nicotinic acetylcholine receptor gene improves cognitive deficits and synaptic pathology in a mouse model of Alzheimer's disease, J. Neurosci. **29**, 8805–8815 (2009)

51.148 A.M. Lilja, O. Porras, E. Storelli, A. Nordberg, A. Marutle: Functional interactions of fibrillar and oligomeric amyloid-β with alpha7 nicotinic receptors in Alzheimer's disease, J. Alzheimers Dis. **23**, 335–347 (2011)

51.149 Y. An, X.L. Qi, J.J. Pei, Z. Tang, Y. Xiao, Z.Z. Guan: Amyloid precursor protein gene mutated at Swedish 670/671 sites in vitro induces changed expression of nicotinic acetylcholine receptors and neurotoxicity, Neurochem. Int. **57**, 647–654 (2010)

51.150 G. Niewiadomska, A. Mietelska-Porowska, M. Mazurkiewicz: The cholinergic system, nerve growth factor and the cytoskeleton, Behav. Brain Res. **221**, 515–526 (2011)

51.151 S. Capsoni, R. Brandi, I. Arisi, M. D'Onofrio, A. Cattaneo: A dual mechanism linking NGF/proNGF imbalance and early inflammation to Alzheimer's disease neurodegeneration in the AD11 Anti-NGF mouse model, CNS Neurol. Disord. Drug Targets, **10**, 635–647 (2011)

51.152 A. Salehi, J.D. Delcroix, P.V. Belichenko, K. Zhan, C. Wu, J.S. Valletta, R. Takimoto-Kimura, A.M. Kleschevnikov, K. Sambamurti, P.P. Chung, W. Xia, A. Villar, W.A. Campbell, L.S. Kulnane, R.A. Nixon, B.T. Lamb, C.J. Epstein, G.B. Stokin, L.S. Goldstein, C. Mobley: Increased APP expression in a mouse model of Down's syndrome disrupts NGF transport and causes cholinergic neuron degeneration, Neuron **51**, 29–42 (2006)

51.153 J. Fombonne, S. Rabizadeh, S. Banwait, P. Mehlen, D.E. Bredesen: Selective vulnerability in Alzheimer's disease: Amyloid precursor protein and p75(NTR) interaction, Ann. Neurol. **65**, 294–303 (2009)

51.154 K.K. Teng, S. Felice, T. Kim, B.L. Hempstead: Understanding proneurotrophin actions: Recent advances and challenges, Dev. Neurobiol. **70**, 350–359 (2010)

51.155 M. Fahnestock, G. Yu, M.D. Coughlin: ProNGF: A neurotrophic or an apoptotic molecule?, Prog. Brain Res. **146**, 101–110 (2004)

51.156 M.A. Bruno, W.C. Leon, G. Fragoso, W.E. Mushynski, G. Almazan, A.C. Cuello: Amyloid β-induced nerve growth factor dysmetabolism in Alzheimer disease, J. Neuropathol. Exp. Neurol. **68**, 857–869 (2009)

51.157 E.J. Coulson, L.M. May, A.M. Sykes, A.S. Hamlin: The role of the p75 neurotrophin receptor in cholinergic dysfunction in Alzheimer's disease, Neuroscientist **15**, 317–322 (2009)

51.158 C. Costantini, F. Rossi, E. Formaggio, R. Bernardoni, D. Cecconi, V. Della-Bianca: Characterization of the signaling pathway downstream p75 neurotrophin receptor involved in β-amyloid peptide-dependent cell death, J. Mol. Neurosci. **25**, 141–156 (2005)

51.159 E.J. Coulson: Does the p75 neurotrophin receptor mediate Aβ-induced toxicity in Alzheimer's disease?, J. Neurochem. **98**, 654–660 (2006)

51.160 A. Sotthibundhu, A.M. Sykes, B. Fox, C.K. Underwood, W. Thangnipon, E.J. Coulson: β-$Amyloid_{1-42}$ induces neuronal death through the p75 neurotrophin receptor, J. Neurosci. **28**, 3941–3946 (2008)

51.161 J.K. Knowles, J. Rajadas, T.V. Nguyen, T. Yang, M.C. LeMieux, L. Vander Griend, C. Ishikawa, S.M. Massa, T. Wyss-Coray, F.M. Longo: The p75 neurotrophin receptor promotes amyloid-beta (1–42)-induced neuritic dystrophy in vitro and in vivo, J. Neurosci. **29**, 10627–10637 (2009)

51.162 M. Yaar, S. Zhai, I. Panova, R.E. Fine, P.B. Eisenhauer, J.K. Blusztajn, I. Lopez-Coviella, B.A. Gilchrest: A cyclic peptide that binds p75(NTR) protects neurones from β amyloid (1–40)-induced cell death, Neuropathol. Appl. Neurobiol. **33**, 533–543 (2007)

51.163 Y.J. Wang, X. Wang, J.J. Lu, Q.X. Li, C.Y. Gao, X.H. Liu, Y. Sun, M. Yang, Y. Lim, G. Evin, J.H. Zhong, C. Masters, X.F. Zhou: p75NTR regulates Aβ deposition by increasing Aβ production but inhibiting Aβ aggregation with its extracellular domain, J. Neurosci. **31**, 2292–2304 (2011)

51.164 B. Chakravarthy, C. Gaudet, M. Ménard, T. Atkinson, L. Brown, F.M. Laferla, U. Armato, J. Whitfield: Amyloid-β peptides stimulate the expression of the p75(NTR) neurotrophin receptor in SHSY5Y human neuroblastoma cells and AD transgenic mice, J. Alzheimers Dis. **19**, 915–925 (2010)

51.165 M. Goedert, R. Jakes: Mutations causing neurodegenerative tauopathies, Biochim. Biophys. Acta **1739**, 240–250 (2005)

51.166 S. Oddo, A. Caccamo, M. Kitazawa, B.P. Tseng, F.M. LaFerla: Amyloid deposition precedes tangle formation in a triple transgenic model of Alzheimer's disease, Neurobiol. Aging **24**, 1063–1070 (2003)

51.167 L.M. Billings, S. Oddo, K.N. Green, J.L. McGaugh, F.M. LaFerla: Intraneuronal Aβ causes the onset of early Alzheimer's disease-related cognitive deficits in transgenic mice, Neuron **45**, 675–688 (2005)

51.168 M. Kitazawa, S. Oddo, T.R. Yamasaki, K.N. Green, F.M. LaFerla: Lipopolysaccharide-induced inflammation exacerbates tau pathology by a cyclin-dependent kinase 5-mediated pathway in a transgenic model of Alzheimer's disease, J. Neurosci. **25**, 8843–8853 (2005)

51.169 S.D. Ginsberg, S. Che, S.E. Counts, E.J. Mufson: Shift in the ratio of three-repeat tau and four-repeat tau mRNAs in individual cholinergic basal forebrain neurons in mild cognitive impairment and Alzheimer's disease, J. Neurochem. **96**, 1401–1408 (2006)

51.170 M. Mesulam: The cholinergic lesion of Alzheimer's disease: Pivotal factor or side show, Learn Mem. **11**, 43–49 (2004)

51.171 C. Geula, N. Nagykery, A. Nicholas, C.K. Wu: Cholinergic neuronal and axonal abnormalities are present early in aging and in Alzheimer disease, J. Neuropathol. Exp. Neurol. **67**, 309–318 (2008)

51.172 K. Belarbi, S. Burnouf, F.J. Fernandez-Gomez, J. Desmerciéres, L. Troquier, J. Brouillette, L. Tsambou, M.E. Grosjean, R. Caillierez, D. Demeyer, M. Hamdane, K. Schindowski, D. Blum, L. Buée: Loss of medial septum cholinergic neurons in THY-tau22 mouse model: What links with tau pathology?, Curr. Alzheimer Res. **8**, 633–638 (2011)

51.173 J.C. de la Torre: Vascular risk factor detection and control may prevent Alzheimer's disease, Ageing Res. Rev. **9**, 218–225 (2010)

51.174 C. Iadecola, L. Park, C. Capone: Threats to the mind: Aging, amyloid, and hypertension, Stroke. **40**(3 Suppl), S40–44 (2009)

51.175 J.C. de la Torre, T. Mussivand: Can disturbed brain microcirculation cause Alzheimer's disease?, Neurol. Res. **15**, 146–153 (1993)

51.176 R.N. Kalaria: Vascular basis for brain degeneration: Faltering controls and risk factors for dementia, Nutr. Rev. **68**(Suppl.), S74–87 (2010)

51.177 N. Nicolakakis, E. Hamel: Neurovascular function in Alzheimer's disease patients and experimental models, Cereb. Blood Flow Metab. **31**, 1354–1370 (2011)

51.178 R.O. Weller, D. Boche, J.A.R. Nicoll: Microvasculature changes and cerebral amyloid angiopathy in Alzheimer's disease and their potential impact on therapy, Acta Neuropathol. **119**, 87–102 (2009)

51.179 C. Cordonnier, W.M. van der Flier: Brain microbleeds and Alzheimer's disease: Innocent observation or key player?, Brain **134**, 335–344 (2011)

51.180 B.V. Zlokovic: New therapeutic targets in the neurovascular pathway in Alzheimer's disease, Neurotherapeutics **5**, 409–414 (2008)

51.181 C.A. Hawkes, W. Härtig, J. Kacza, R. Schliebs, R.O. Weller, J.A. Nicoll, R.O. Carare: Perivascular drainage of solutes is impaired in the ageing mouse brain and in the presence of cerebral amyloid angiopathy, Acta Neuropathol. **121**, 431–443 (2011)

51.182 M. Merlini, E.P. Meyer, A. Ulmann-Schuler, R.M. Nitsch: Vascular β-amyloid and early astrocyte alterations impair cerebrovascular function and cerebral metabolism in transgenic arcAβ mice, Acta Neuropathol. **122**, 293–311 (2011)

51.183 D. Paris, J. Humphrey, A. Quadros, N. Patel, R. Crescentini, F. Crawford, M. Mullan: Vasoactive effects of Aβ in isolated human cerebrovessels and in a transgenic mouse model of Alzheimer's disease: Role of inflammation, Neurol. Res. **25**, 642–651 (2003)

51.184 C. Iadecola: Neurovascular regulation in the normal brain and in Alzheimer's disease, Nat Rev. Neurosci. **5**, 347–360 (2004)

51.185 A. Rocchi, D. Orsucci, G. Tognoni, R. Ceravolo, G. Siciliano: The role of vascular factors in late-onset sporadic Alzheimer's disease. Genetic and molecular aspects, Curr. Alzheimer Res. **6**, 224–237 (2009)

51.186 H.J. Milionis, M. Florentin, S. Giannopoulos: Metabolic syndrome and Alzheimer's disease: A link to a vascular hypothesis?, CNS Spectrums **13**, 606–613 (2008)

51.187 D.A. Snowdon, L.H. Greiner, J.A. Mortimer, K.P. Riley, P.A. Greiner, W.R. Markesbery: Brain infarction and the clinical expression of Alzheimer disease, The Nun Study, J. Am. Med. Assoc. **277**, 813–817 (1997)

51.188 S.L. Cole, R. Vassar: Linking vascular disorders and Alzheimer's disease: Potential involvement of BACE1, Neurbiol. Aging **30**, 1535–1544 (2009)

51.189 E.E. Smith, S.M. Greenberg: β-amyloid, blood vessels and brain function, Stroke **40**, 2601–2606 (2009)

51.190 H.K. Shin, P.B. Jones, M. Garcia-Alloza, L. Borrelli, S.M. Greenberg, B.J. Bacskai, M.P. Frosch, B.T. Hyman, M.A. Moskowitz, C. Ayata: Age-dependent cerebrovascular dysfunction in a transgenic mouse model of cerebral amyloid angiopathy, Brain. **130**(Pt 9), 2310–2319 (2007)

51.191 D. Paris, N. Patel, A. DelleDonne, A. Quadros, R. Smeed, M. Mullan: Impaired angiogenesis in a transgenic mouse model of cerebral amyloidosis, Neurosci. Lett. **366**, 80–85 (2004)

51.192 D. Paris, K. Townsend, A. Quadros, J. Humphrey, J. Sun, S. Brem, M. Wotoczek-Obadia, A. DelleDonne, N. Patel, D.F. Obregon, R. Crescentini, L. Abdullah, D. Coppola, A.M. Rojiani, F. Crawford, S.M. Sebti, M. Mullan: Inhibition of angiogenesis by Aβ peptides, Angiogenesis **7**, 75–85 (2004)

51.193 C. Ruiz de Almodovar, D. Lambrechts, M. Mazzone, P. Carmeliet: Role and therapeutic potential of VEGF in the nervous system, Physiol. Rev. **89**, 607–648 (2009)

51.194 F.Y. Sun, X. Guo: Molecular and cellular mechanisms of neuroprotection by vascular endothelial growth factor, J. Neurosci. Res. **79**, 180–184 (2005)

51.195 N. Ferrara, H.P. Gerber, J. LeCouter: The biology of VEGF and its receptors, Nat. Med. **9**, 669–676 (2003)

51.196 R.N. Kalaria, D.L. Cohen, D.R. Premkumar, S. Nag, J.C. LaManna, W.D. Lust: Vascular endothelial growth factor in Alzheimer's disease and experimental cerebral ischemia, Mol. Brain Res. **62**, 101–105 (1998)

51.197 E. Tarkowski, R. Issa, M. Sjogren, A. Wallin, K. Blennow, A. Tarkowski: Increased intrathecal levels of the angiogenic factors VEGF and TGF-beta in Alzheimer's disease and vascular dementia, Neurobiol. Aging **23**, 237–243 (2002)

51.198 S.P. Yang, D.G. Bae, H.J. Kang, B.J. Gwag, Y.S. Gho, C.B. Chae: Co-accumulation of vascular endothelial growth factor with β-amyloid in the brain of patients with Alzheimer's disease, Neurobiol. Aging **25**, 283–290 (2004)

51.199 J.K. Ryu, T. Cho, H.B. Choi, Y.T. Wang, J.G. McLarnon: Microglial VEGF receptor response is an integral chemotactic component in Alzheimer's disease pathology, J. Neurosci. **29**, 3–13 (2009)

51.200 A.I. Pogue, W.J. Lukiw: Angiogenic signaling in Alzheimer's disease, Neuroreport **15**, 1507–1510 (2004)

51.201 L. Thirumangalakudi, P.G. Samany, A. Owoso, B. Wiskar, P. Grammas: Angiogenic proteins are expressed by brain blood vessels in Alzheimer's disease, J. Alzheimer Dis. **10**, 111–118 (2006)

51.202 A.H. Vagnucci, W.W. Li: Alzheimer's disease and angiogenesis, Lancet **361**, 605–608 (2003)

51.203 B.S. Desai, J.A. Schneider, J.L. Li, P.M. Carvey, B. Hendey: Evidence of angiogenic vessels in Alzheimer's disease, J. Neural Transm. **116**, 587–597 (2009)

51.204 H.J. Marti, M. Bernaudin, A. Bellail, H. Schoch, M. Euler, W. Petit: Hypoxia-induced vascular endothelial growth factor expression precedes neovascularization after cerebral ischemia, Am. J. Pathol. **156**, 965–976 (2000)

51.205 I. Stein, M. Neeman, D. Shweiki, A. Itin, E. Keshet: Stabilization of vascular endothelial growth factor mRNA by hypoxia and hypoglycemia and coregulation with other ischemia-induced genes, Mol. Cell Biol. **15**, 5363–5368 (1995)

51.206 G.D. Yancopoulos, S. Davis, N.W. Gale, J.S. Rudge, S.J. Wiegand, J. Holash: Vascular-specific growth factors and blood vessel formation, Nature **407**, 242–248 (2000)

51.207 D. Paris, A. Quadros, N. Patel, A. DelleDonne, J. Humphrey, M. Mullan: Inhibition of angiogenesis and tumour growth by β- and γ-secretase inhibitors, Eur. J. Pharmacol. **514**, 1–15 (2005)

51.208 J.R. Ciallella, H. Figueiredo, V. Smith-Swintosky, J.P. McGillis: Thrombin induces surface and intracellular secretion of amyloid precursor protein from human endothelial cells, Thromb. Haemost. **81**, 630–637 (1999)

51.209 S. Bürger, M. Noack, L.P. Kirazov, E.P. Kirazov, C.L. Naydenov, E. Kouznetsova, Y. Yafai, R. Schliebs: Vascular endothelial growth factor (VEGF) affects processing of the amyloid precursor protein and β-amyloidogenesis in brain slice cultures derived from transgenic Tg2576 mouse brain, Int. J. Dev. Neurosci. **27**, 517–523 (2009)

51.210 S. Bürger, Y. Yafai, M. Bigl, P. Wiedemann, R. Schliebs: Effect of VEGF and its receptor antagonist SU-5416, an inhibitor of angiogenesis, on processing of the β-amyloid precursor protein in primary neuronal cells derived from brain tissue of Tg2576 mice, Int. J. Dev. Neurosci. **28**, 597–604 (2010)

51.211 H. Girouard, C. Iadecola: Neurovascular coupling in the normal brain and in hypertension, stroke, and Alzheimer disease, J. Appl. Physiol. **100**, 328–335 (2006)

51.212 E. Hamel: Perivascular nerves and the regulation of cerebrovascular tone, J. Appl. Physiol. **100**, 1059–1064 (2006)

51.213 A.H. VanBeek, J.A. Claassen: The cerebrovascular role of the cholinergic neural system in Alzheimer's disease, Behav. Brain Res. **221**, 537–542 (2003)

51.214 E. Vaucher, E. Hamel: Cholinergic basal forebrain neurons project to cortical microvessels in the rat: Electron microscopic study with anterogradely transported *Phaseolus vulgaris* leucoagglutinin and choline acetyltransferase immunocytochemistry, J. Neurosci. **15**, 7427–7441 (1995)

51.215 E. Hamel: Cholinergic modulation of the cortical microvascular bed, Prog. Brain Res. **145**, 171–178 (2004)

51.216 A. Elhusseiny, E. Hamel: Muscarinic–but not nicotinic–acetylcholine receptors mediate a nitric oxide-dependent dilation in brain cortical arterioles: A possible role for the M5 receptor subtype, J. Cereb. Blood Flow Metab. **20**, 298–305 (2000)

51.217 E. Farkas, P.G. Luiten: Cerebral microvascular pathology in aging and Alzheimer's disease, Prog. Neurobiol. **64**, 575–611 (2001)

51.218 B. Cauli, X.K. Tong, A. Rancillac, N. Serluca, B. Lambolez, J. Rossier, E. Hamel: Cortical GABA interneurons in neurovascular coupling: Relays for subcortical vasoactive pathways, J. Neurosci. **24**, 8940–8949 (2004)

51.219 C. Humpel, J. Marksteiner: Cerebrovascular damage as a cause for Alzheimer's disease, Curr. Neurovasc. Res. **2**, 341–347 (2005)

51.220 J.A. Claassen, R.W. Jansen: Cholinergically mediated augmentation of cerebral perfusion in Alzheimer's disease and related cognitive disorders: The cholinergic-vascular hypothesis, J. Gerontol. A Biol. Sci. Med. Sci. **61**, 267–271 (2006)

51.221 E. Kouznetsova, R. Schliebs: Role of cholinergic system in β-amyloid related changes of perivascular innervation of cerebral microvessels in transgenic Tg2576 Alzheimer-like mice, J. Neurochem. **101**(Suppl. 1), 63 (2007)

51.222 A. Szutowicz, H. Bielarczyk, S. Gul, P. Zieliński, T. Pawełczyk, M. Tomaszewicz: Nerve growth factor and acetyl-l-carnitine evoked shifts in acetyl-CoA

and cholinergic SN56 cell Vulnerability to neurotoxic inputs, J. Neurosci. Res. **79**, 185–192 (2005)
51.223 S.C. Correia, R.X. Santos, G. Perry, X. Zhu, P.I. Moreira, M.A. Smith: Insulin-resistant brain state: The culprit in sporadic Alzheimer's disease?, Ageing Res. Rev. **10**, 264–273 (2011)
51.224 M. Bigl, J. Apelt, K. Eschrich, R. Schliebs: Cortical glucose metabolism is altered in aged transgenic Tg2576 mice that demonstrate Alzheimer plaque pathology, J. Neural Transm. **110**, 77–94 (2003)
51.225 P. Ghosh, A. Kumar, B. Datta, V. Rangachari: Dynamics of protofibril elongation and association involved in Aβ42 peptide aggregation in Alzheimer's disease, BMC Bioinformatics **11**(Suppl 6), S24 (2010)
51.226 N.L. Fawzi, E.H. Yap, Y. Okabe, K.L. Kohlstedt, S.P. Brown, T. Head-Gordon: Contrasting disease and nondisease protein aggregation by molecular simulation, Acc. Chem. Res. **41**, 1037–1047 (2008)
51.227 A.R. George, D.R. Howlett: Computationally derived structural models of the beta-amyloid found in Alzheimer's disease plaques and the interaction with possible aggregation inhibitors, Biopolymers **50**, 733–741 (1999)
51.228 S.Y. Ponomarev, J. Audie: Computational prediction and analysis of the DR6-NAPP interaction, Proteins. **79**, 1376–1395 (2011)
51.229 J. Luo, J.D. Maréchal, S. Wärmländer, A. Gräslund, A. Perálvarez-Marín: In silico analysis of the apolipoprotein E and the amyloid beta peptide interaction: Misfolding induced by frustration of the salt bridge network, PLoS Comput. Biol. **6**(2), e1000663 (2010)
51.230 J.Y. Chen, E. Youn, S.D. Mooney: Connecting protein interaction data, mutations, Methods Mol. Biol. **541**, 449–461 (2009)
51.231 K.C. Chou, K.D. Watenpaugh, R.L. Heinrikson: A model of the complex between cyclin-dependent kinase 5 and the activation domain of neuronal Cdk5 activator, Biochem. Biophys. Res. Commun. **259**, 420–428 (1999)
51.232 Y. Huang, X. Sun, G. Hu: An integrated genetics approach for identifying protein signal pathways of Alzheimer's disease, Comput. Methods Biomech. Biomed. Engin. **14**, 371–378 (2011)
51.233 D.L. Cook, J.C. Wiley, J.H. Gennari: Chalkboard: Ontology-based pathway modeling and qualitative inference of disease mechanisms, Pac. Symp. Biocomput. **2007**, 16–27 (2007)
51.234 I. García, Y. Fall, G. Gómez, H. González-Díaz: First computational chemistry multi-target model for anti-Alzheimer, anti-parasitic, anti-fungi, and anti-bacterial activity of GSK-3 inhibitors in vitro, in vivo, and in different cellular lines, Mol. Divers. **15**, 561–567 (2011)
51.235 R. Gómez-Balderas, D.F. Raffa, G.A. Rickard, P. Brunelle, A. Rauk: Computational studies of Cu(II)/Met and Cu(I)/Met binding motifs relevant for the chemistry of Alzheimer's disease, J. Phys. Chem. A **109**, 5498–5508 (2005)
51.236 R. Singh, A. Barman, R. Prabhakar: Computational insights into aspartyl protease activity of presenilin 1 (PS1) generating Alzheimer amyloid β-peptides (Aβ40 and Aβ42), J. Phys. Chem. B. **113**, 2990–2999 (2009)
51.237 M. Eisenstein: Genetics: Finding risk factors, Nature **475**, S20–S22 (2011)
51.238 M.D. Ritchie, L.W. Hahn, J.H. Moore: Power of multifactor dimensionality reduction for detecting gene–gene interactions in the presence of genotyping error, missing data, phenocopy, and genetic heterogeneity, Genet. Epidemiol. **24**, 150–157 (2003)
51.239 S.Y. Lee, Y. Chung, R.C. Elston, Y. Kim, T. Park: Log-linear model-based multifactor dimensionality reduction method to detect gene gene interactions, Bioinformatics. **23**, 2589–2595 (2007)
51.240 J. Andrade, M. Andersen, A. Sillén, C. Graff, J. Odeberg: The use of grid computing to drive data-intensive genetic research, Eur. J. Hum. Genet. **15**, 694–702 (2007)
51.241 B. Liu, T. Jiang, S. Ma, H. Zhao, J. Li, X. Jiang, J. Zhang: Exploring candidate genes for human brain diseases from a brain-specific gene network, Biochem. Biophys. Res. Commun. **349**, 1308–1314 (2006)
51.242 K. Heinitz, M. Beck, R. Schliebs, J.R. Perez-Polo: Toxicity mediated by soluble oligomers of β-amyloid (1–42) on cholinergic SN56.B5.G4 cells, J. Neurochem. **98**, 1930–1945 (2006)
51.243 S. Joerchel, M. Raap, M. Bigl, K. Eschrich, R. Schliebs: Oligomeric β-amyloid (1–42) induces the expression of Alzheimer disease-relevant proteins in cholinergic SN56.B5.G4 cells as revealed by proteomic analysis, Int. J. Dev. Neurosci. **26**, 301–308 (2006)
51.244 J. Li, X. Zhu, J.Y. Chen: Building disease-specific drug-protein connectivity maps from molecular interaction networks and PubMed abstracts, PLoS Comput. Biol. **5**(7), e1000450 (2009)
51.245 Y. He, Z. Chen, G. Gong, A. Evans: Neuronal networks in Alzheimer's disease, Neuroscientist **15**, 333–350 (2009)
51.246 D.J. Watts, S.H. Strogatz: Collective dynamics of 'small-world' networks, Nature **393**, 440–442 (1998)
51.247 C.J. Stam, B.F. Jones, G. Nolte, M. Breakspear, P. Scheltens: Small-world networks and functional connectivity in Alzheimer's disease, Cereb. Cortex **17**, 92–99 (2007)
51.248 R. Tandon, S. Adak, J.A. Kaye: Neural networks for longitudinal studies in Alzheimer's disease, Artif. Intell. Med. **36**, 245–255 (2006)
51.249 M.E. Hasselmo: A computational model of the progression of Alzheimer's disease, M.D. Computing **14**, 181–191 (1997)

51.250 M.E. Hasselmo: Neuromodulation and cortical function: Modeling the physiological basis of behavior, Behav. Brain Res. **67**, 1–27 (1995)
51.251 D. Horn, N. Levy, E. Ruppin: Neuronal-based synaptic compensation: A computational study in Alzheimer's disease, Neural Comput. **8**, 1227–1243 (1996)
51.252 L. Nickels, B. Biedermann, M. Coltheart, S. Saunders, J.J. Tree: Computational modelling of phonological dyslexia: How does the DRC model fare?, Cogn. Neuropsychol. **25**, 165–193 (2008)
51.253 A. Serna, V. Rialle, H. Pigot: Computational representation of Alzheimer's disease evolution applied to a cooking activity, Stud. Health Technol. Inform. **124**, 587–592 (2006)
51.254 M.A. Ahmadi-Pajouh, F. Towhidkhah, S. Gharibzadeh, M. Mashhadimalek: Path planning in the hippocampo-prefrontal cortex pathway: An adaptive model based receding horizon planner, Med. Hypotheses **68**, 1411–1415 (2007)
51.255 E. Ruppin, J.A. Reggia: A neural model of memory impairment in diffuse cerebral atrophy, Br. J. Psychiatry. **166**, 19–28 (1995)