

# Chapter 12

## Possible Role of Resveratrol Targeting Estradiol and Neprilysin Pathways in Lipopolysaccharide Model of Alzheimer Disease

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**Abstract** Alzheimer's disease (AD) is an irreversible, progressive neurodegenerative brain disease that slowly destroys memory and thinking skills. It is the most common cause of dementia among older people. One of the most important hallmarks of AD is the presence of amyloid beta ( $A\beta$ ) peptide in the brain that suggests that it is the primary trigger for neuronal loss. Herbal extracts have been studied over the years for their potential therapeutic effect in AD. Resveratrol (RSV), one of the most important phytoestrogens, is considered to be useful as estrogen plays an important role in AD. One of the most important amyloid degrading enzymes is neprilysin (NEP), which plays a major role in degrading  $A\beta$ , and mainly affected by estrogen. So, the aim of the present study is investigating the possible role of resveratrol in lipopolysaccharide model of AD and the implication of its possible role in regulating the estradiol and neprilysin pathways. Mice were divided into four groups: Control group (0.9 % saline), LPS group (0.8 mg/kg i.p once), Treatment group with RSV (mice were once injected with LPS then after 30 min given a dose of { 4 mg/kg } RSV for 7 days), and RSV group only (mice received 4 mg/kg i.p for 7 days only). After 7 days mice were subjected to different behavioral tests using Y-maze, object recognition test, and open field tests. Estradiol and NEP level were measured using ELISA kit. Results showed RSV was able to reverse the decline in different types of memory (working, nonspatial, and locomotor functions) caused by LPS induction in mice. Moreover RSV was able to significantly increase both the estradiol level and NEP level and that may have a great role to decrease  $A\beta$  deposition as it has been confirmed that there is a link between NEP and estradiol

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level; by upregulation of estradiol level this consequently leads to increase in the level of NEP level, and by increasing the NEP level in brain, this lead to decrease in A $\beta$  deposition and enhancing its degradation by NEP

**Keywords** Alzheimer disease • Resveratrol • Neprilysin • Lipopolysaccharide • Estrogen

## Abbreviations

(A $\beta$ )	Amyloid beta peptides
AD	Alzheimer's disease
DES	Diethylstilbestrol
DR	Discrimination ratio
i.p.	Intraperitoneal
LPS	Lipopolysaccharide
MAP	Mean alternation percentage
NEP	Neprilysin
RSV	Resveratrol

## 12.1 Introduction

Alzheimer's disease (AD) is a progressive neurodegenerative disease that is characterized by impairment of memory and disturbance in at least one of the thinking functions. It is considered the most common cause of dementia, affecting more than 25 million people worldwide [5] and considered the fifth leading cause of death for those aged more than 65 years. The most common hypothesis behind AD is the deposition of neurotic plaques extracellularly and neurofibrillary tangles intracellularly in the brain of AD patients. Several major theories are involved in the pathogenesis of AD, such as neurotic plaques hypothesis [45], tauopathy [30], inflammation [37], and oxidative stress [41].

Neurotic plaque hypothesis is considered the primary hallmark of AD in which APP (protein normally found in brain essential for the growth and survival of the neurons) is cleaved by mutant form of  $\beta$ -secretase then followed by  $\gamma$ -secretase which results in generation of the A $\beta$  peptide which aggregates and deposit on the surface of the neurons [32].

Different mechanisms were implemented in order to decrease the level of beta amyloid plaques, one of them is altering catabolism of A $\beta$  levels in the brains of AD, by using many proteases and peptidases that have the capability of cleaving A $\beta$  either in vitro or in vivo. The following proteinases have the abilities of degrading A $\beta$  peptide. One of the primary degrading enzyme which targets the degradation of beta amyloid peptides is Neprilysin enzyme (NEP) which is found to be abundant in the brain areas vulnerable to amyloid plaque deposition [23]. It is considered the major amyloid degrading enzyme and plays a major role in the clearance of amyloid

beta peptides ( $A\beta$ ) from the brain [22, 36]. NEP was found to degrade both monomeric and oligomeric forms of  $A\beta_{40}$  and  $A\beta_{42}$  in intracellular and extracellular compartments of the brain [16, 56].

Estrogen deprivation has been implicated in the pathogenesis of AD [54]. Furthermore, there is a strong evidence that AD pathology and AD-related cognitive decline are strongly related to decrease in estrogen level [43, 53]. Estrogen plays an important role in protection against neuron cell death [48, 52] and has the ability of inhibition of different aspects of AD neuropathology including  $A\beta$  accumulation [4, 42].

Interestingly NEP was found to be mainly upregulated by estrogen hormone [19]. Moreover it has been proven that estrogen deprivation caused a lowering of NEP level and that estrogen replacement therapy restored NEP level to control values in ovariectomized rat brains. It has been speculated that estrogen regulates the expression of NEP [13]. Furthermore NEP activity was found to be lower in the hippocampus, cerebellum, and caudate of ovariectomized rats than in non-ovariectomized rats, and this effect can be reversed by exogenous  $17\beta$ -estradiol [59].

Lipopolysaccharide (LPS) is a gram negative bacterial endotoxin that was found to induce neuroinflammation which exhibits AD-like features when systemically injected to rodents [7]. Moreover it has been proven that intraperitoneal (i.p.) injection of LPS induces cognitive impairment in mice as it induces memory loss and amyloidogenesis in vivo and in vitro as a consequence of systemic inflammation [46, 51] by production of proinflammatory cytokines such as IL-1 $\beta$ , IL-6, and TNF- $\alpha$  [38, 17].

Resveratrol (RSV) (3,4',5-trihydroxystilbene), a secondary metabolite, belongs to a class of polyphenolic compounds called stilbenes [49]. It was first isolated from the roots of *Veratrum grandiflorum* [29]. Several plant species are known to produce RSV (classified as polyphenol) such as vine plant, peanuts, berries but was found to be in large amounts in skin of red grapes [11, 40].

RSV possesses many biological activities that are protective against atherosclerosis, including antioxidant, anti-inflammatory, antiplatelet, and vasorelaxant activities, and has been used in applying cardioprotective effect [35]. In addition, it is identified as a cancer chemotherapeutic agent [26] and has recently been reported to mimic effects of dietary restriction and extends the life span of lower organisms [18, 58].

It has been demonstrated that RSV can lessen amyloid- $\beta$  peptide-induced toxicity, protect against cerebral ischemic injury, and shield neurons from induced excitotoxicity [28]. Moreover, RSV promotes the non-amyloidogenic pathway and enhances the cleavage of the amyloid precursor protein. Furthermore it enhances clearance of amyloid beta peptides, and reduces neuronal damage [31].

One of the important features that RSV has been characterized with is being a phytoestrogen on the basis of its ability to bind to estrogen receptors  $\alpha$  and  $\beta$  (ERs  $\alpha$  and ER  $\beta$ ) and elicits similar responses to endogenous estrogens. Moreover, the chemical structure of RSV is very similar to that of the synthetic estrogen agonist, diethylstilbestrol (DES). RSV is considered an estrogen agonist [6]. It was found

that RSV in particular acts on estrogen receptor beta which has a major function in the cognitive processes, so by binding of RSV to estrogen receptor beta, this results in improvement of cognitive impairment which results from AD [10].

So due to the structural similarity between RSV and estradiol this study was conducted to investigate the strong correlation between RSV and NEP in the degradation of amyloid beta.

## 12.2 Methodology

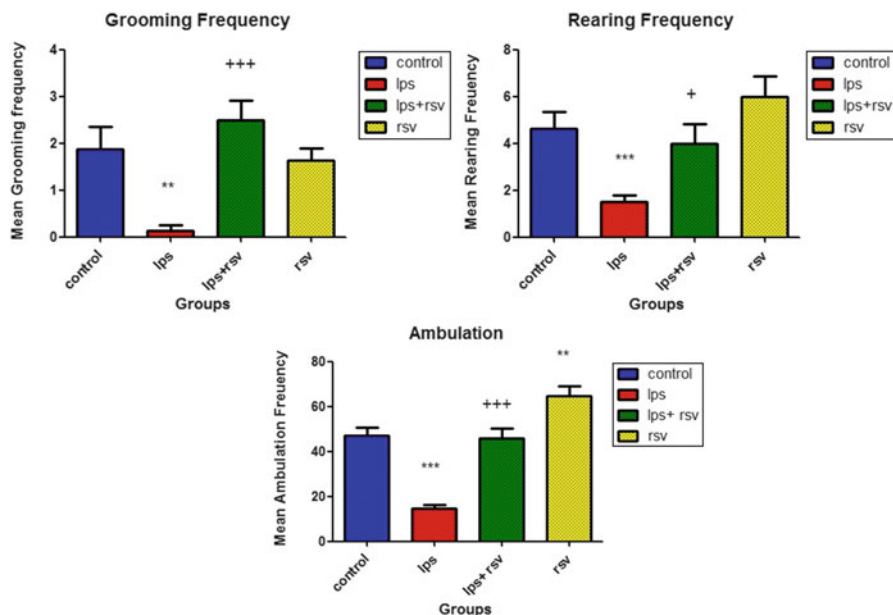
After 1 week of acclimatization, mice were randomly allocated to four groups each consisting of 10–12 mice. One group served as normal and received (saline 0.9 % i.p.) once.

The second group mice were injected with LPS (0.8 mg/kg i.p in saline) once to induce systemic and neuroinflammation and then were subjected to the tests after 7 days [1, 47, 14]. The third group mice were injected with LPS (0.8 mg/kg i.p in saline) once, and after 30 min mice were injected with RSV (4 mg/kg in saline) for 7 days [8, 9]. RSV was dissolved in the least amount of DMSO and completed to the final volume with saline [12, 24]. Animals were subjected to the tests after the seventh day of all injections. The fourth group mice were injected with only RSV (4 mg/kg i.p in saline) and were subjected to the tests after the seventh day of injections. All Animals were subjected to different behavioral tests such as open field, object recognition, and Y-maze behavioral tests at the end of the previously mentioned treatments, then they were sacrificed, and their brains were harvested. The brains were divided into two hemispheres and were frozen at 80 °C to be later homogenized and used for ELISA Kit.

## 12.3 Relation Between Resveratrol as a Phytoestrogen and Neprilysin in Lipopolysaccharide Induced Alzheimer's Disease

Behavioral tests were performed as a form of noninvasive method to detect changes in cognitive function after treating mice with LPS. Three behavioral tests were conducted: Y-maze test to detect working memory, open field test to measure locomotor functions, and object recognition test to measure nonspatial memory.

LPS-induced cognitive impairment was associated with a decrease in ambulation, rearing, and grooming frequencies compared to the normal animals in open field testing. It also resulted in significant decrease in working memory evaluated by significant decrease in MAP measured by Y-maze test. These results are in agreement with those of [14], in which administration of LPS once then testing



**Fig. 12.1** Effects of LPS and resveratrol on the ambulation, grooming, and rearing frequencies

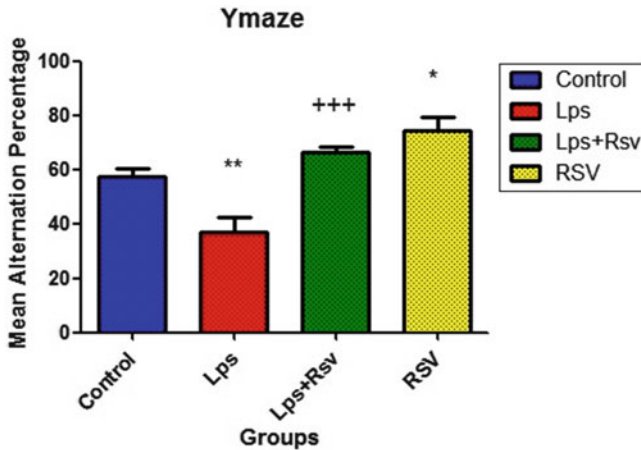
mice after 7 days from injection significantly reduced locomotor activity as well as reduced MAP (Figs. 12.1 and 12.2).

LPS-induced AD was associated with a decrease in nonspatial memory evaluated using new object recognition test. The results showed a significant decrease in the discrimination ratio compared to normal mice. This result is in agreement with those of [25] in which LPS injection showed significant nonspatial memory deficit evaluated by the impairment of object memory in new object recognition test. This object recognition memory deficit was evaluated to be due to systemic response induced by the i.p. injection of LPS (Fig. 12.3).

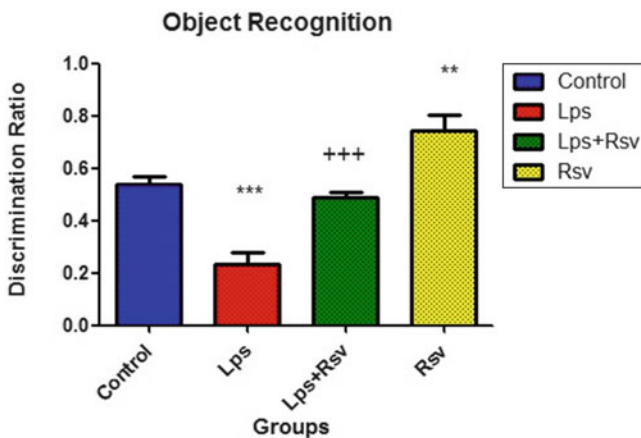
RSV, a polyphenolic compound found mainly in skin of red grapes, was found to induce *in vivo* protective properties against multiple illnesses, including cancer, cardiovascular disease, and ischemia, and was also found to confer resistance to stress and to extend life span [3].

Administration of RSV resulted in significant increase in locomotor activity evaluated by open field test. Our results are in agreement with a study in which RSV has showed that it can contribute to the preservation of cognitive function during aging as it was shown in the results that RSV was able to improve the locomotor function using open field test [44] (Fig. 12.1).

Administration of RSV resulted in significant increase in mean alternation percentage evaluated by Y-maze test. This result is in accordance with a study in which RSV was examined to see its effect on learning and working memory in Y-maze in diabetic rat; results showed that RSV elevated MAP significantly and improved learning and working memory [39] (Fig. 12.2).



**Fig. 12.2** Effect of LPS and resveratrol on the mean alternation percentage

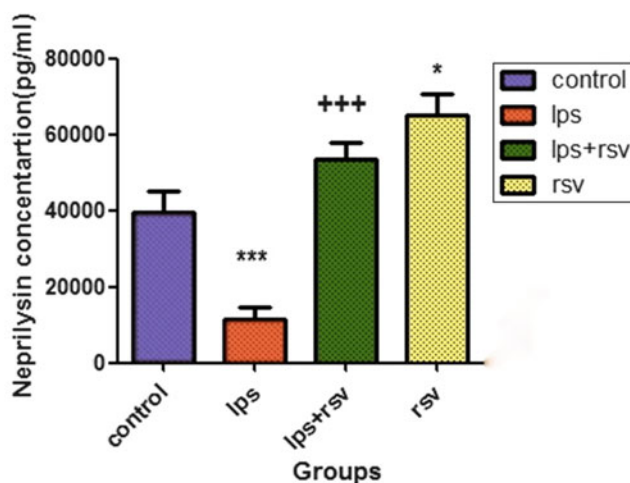
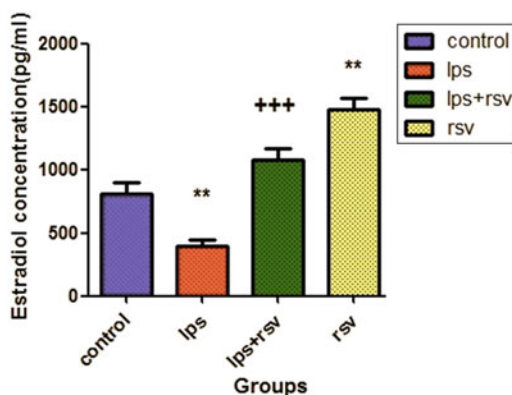


**Fig. 12.3** Effect of LPS and resveratrol on the discrimination ratio

Administration of RSV resulted in significant increase in nonspatial memory evaluated by object recognition test. This result is in harmony with a study in which RSV has been administrated to assess its effect on subarachnoid and intracranial hemorrhage, ischemia, and stroke. Results showed that administration of RSV was able to improve nonspatial memory dramatically after trauma and that posttraumatic memory was restored to 66 % compared to uninjured control [50] (Fig. 12.3).

Previous studies proved that estrogen treatment inhibits microglial activation following exposure to the inflammatory stimuli (LPS) in LPS-treated mice, and that resulted in increasing cell viability and attenuating the inflammatory effect of LPS [55].

**Fig. 12.4** Effect of LPS and resveratrol on estradiol level



**Fig. 12.5** Effect of LPS and resveratrol on neprilysin level

Administration of LPS (0.8 mg/kg i.p.) once significantly decreased estrogen level when measured by estradiol ELISA kit. These results are in agreement with a study done by [57] in which LPS and TNF- $\alpha$  suppress ovarian cell function and decreased estrogen level significantly (Fig. 12.4).

Administration of LPS (0.8 mg/kg i.p.) once significantly decreased NEP level when measured by NEP ELISA kit. These results are in agreement with a study done by [15] in which administration of LPS resulted in significant decrease in the level of NEP (Fig. 12.5).

RSV, a phytoestrogen, has been proven to be able to confer a significant improvement in spatial memory, and protect animals from A $\beta$ -induced neurotoxicity. These neurological protection effects of resveratrol were associated with elevated estrogen level together with reduced iNOS level and oxidative stress [20].

Administration of RSV a phytoestrogen (4 mg/kg i.p) for 7 days either in the group of mice that were injected with LPS to induce AD or in the other group that was injected with RSV, both groups showed a very high significant increase in estrogen level when measured by estradiol ELISA kit based on its structural similarity to diethylstilbestrol. Similar to our results, different studies were conducted to prove that administration of RSV led to elevation in the level of estradiol [2, 27] (Fig. 12.4).

NEP is a zinc metalloendopeptidase that regulates the activity of a number of physiological peptides. Evidence has accumulated that NEP is involved in the clearance of amyloid beta peptides in the brain. Several studies provide evidence that show a strong relation between estrogen and NEP enzyme which is considered the major degrading enzyme for beta amyloid plaques in AD brain. Previous studies have shown that NEP gene responds to progesterone, androgen, and glucocorticoids, but primarily its activity is regulated by estrogen [21].

It has been proven that experimental depletion of endogenous NEP activity by pharmacological inhibitors or reduction of NEP expression by molecular approaches results in neural accumulation of A $\beta$  [33].

Administration of RSV, a phytoestrogen, resulted in significant increase in NEP level when measured by NEP ELISA kit. These results are in agreement with a study by Marambaud et al. where RSV promoted the clearance of Alzheimer's disease amyloid beta peptides through upregulation of NEP level [34] (Fig. 12.5).

## 12.4 Results

Normal animals group were injected once with saline 7 days before subjecting them to open field testing (Group 1). Another group (4) was injected only with RSV (4 mg/kg) for 7 days and was subjected to open field after 7 days. Alzheimer's disease was induced in groups 2 and 3 by intraperitoneal injection of LPS (0.8 mg/kg) 7 days before subjecting them to open field testing. Group 3 was injected with RSV (4 mg/kg i.p.) for 7 days after 30 min from injection the LPS, and was subjected to open field test on the seventh day. Analysis was carried out using unpaired *t*-test to compare each two groups.

Each value represents Mean  $\pm$  Standard Error of Mean. \*Significantly different from Group 1 at  $P < 0.05$ . +Significantly different from Group 2 at  $P < 0.05$ .

Normal animals group were injected once with saline 7 days before subjecting them to Y-maze testing (Group 1). Another group (4) was injected only with RSV (4 mg/kg) for 7 days and was subjected to Y-maze test after 7 days. Alzheimer's disease was induced in groups 2 and 3 by intraperitoneal injection of LPS (0.8 mg/kg) 7 days before subjecting them to Y-maze test. Group 3 was injected with RSV (4 mg/kg) for 7 days after 30 min from injection the LPS, and was subjected to Y-maze test on the seventh day. Analysis was carried out using unpaired *t*-test to compare each two groups.

Each value represents Mean  $\pm$  Standard Error of Mean. \*Significantly different from Group 1 at  $P < 0.05$ . +Significantly different from Group 2 at  $P < 0.05$ .



*MAP* = Mean Alternation Percentage.

Normal animals group were injected once with saline 7 days before subjecting them to object recognition testing (Group 1). Another group (4) was injected only with RSV (4 mg/kg) for 7 days and was subjected to object recognition test after 7 days. Alzheimer's disease was induced in groups 2 and 3 by intraperitoneal injection of LPS (0.8 mg/kg) 7 days before subjecting them to object recognition test. Group 3 was injected with RSV (4 mg/kg) for 7 days after 30 min from injection the LPS, and was subjected to object recognition test on the seventh day. Analysis was carried out using unpaired *t*-test to compare each two groups.

Each value represents Mean  $\pm$  Standard Error of Mean. \*Significantly different from Group 1 at  $P < 0.05$ . <sup>+</sup>Significantly different from Group 2 at  $P < 0.05$ .

*Discrimination Ratio* = DR

Animals were divided into four groups; group1 is the normal animal group injected only with 0.9 % saline i.p once. Alzheimer's disease was induced in groups 2 and 3 by intraperitoneal injection of LPS (0.8 mg/kg i.p) once. Group 2 is the untreated group while the treated group is (group 3) that was injected with RSV (4 mg/kg i.p) for 7 days after 30 min from injection with LPS. Another group of mice injected with RSV only (4 mg/kg i.p) for 7 days (group 4). After performing the behavioral tests, brains were obtained and homogenized as 10 % homogenate in PBS, centrifuged, and supernatants were obtained. Fifty microliters of the supernatants from six samples of each group was diluted to 50  $\mu$ l with assay buffer, added on it reaction mix and assayed. Then substituted by the absorbance in the straight line equation to get the concentrations and calculated as Sa/SV. Statistical analysis of estradiol concentrations was carried out using unpaired *t*-test to compare each two groups.

Each value represents Mean  $\pm$  Standard Error of Mean. \*Significantly different from Group 1 at  $P < 0.05$ . <sup>+</sup>Significantly different from Group 2 at  $P < 0.05$ .

Animals were divided into four groups; group1 is the normal animal group injected only with 0.9 % saline i.p. once. Alzheimer's disease was induced in groups 2 and 3 by intraperitoneal injection of LPS (0.8 mg/kg i.p) once. Group 2 is the untreated group while the treated group is (group 3) that was injected with RSV (4 mg/kg i.p) for 7 days after 30 min from injection with LPS. Another group of mice injected with RSV only (4 mg/kg i.p) for 7 days (group 4). After performing the behavioral tests, brains were obtained and homogenized as 10 % homogenate in PBS, centrifuged, and supernatants were obtained. Fifty microliters of the supernatants from six samples of each group was diluted to 50  $\mu$ l with assay buffer, added on it reaction mix and assayed. Then substituted by the absorbance in the straight line equation to get the concentrations and calculated as Sa/SV. Statistical analysis of estradiol concentrations was carried out using unpaired *t*-test to compare each two groups.

Each value represents Mean  $\pm$  Standard Error of Mean. \*Significantly different from Group 1 at  $P < 0.05$ . <sup>+</sup>Significantly different from Group 2 at  $P < 0.05$ .

### Conclusion

- LPS can be a perfect model for induction of AD in rodents.
- RSV can be a promising drug in management of AD.

### References

1. Arai K et al (2001) Deterioration of spatial learning performances in lipopolysaccharide-treated mice. *Jpn J Pharmacol* 87(3):195–201
2. Basly JP et al (2000) Estrogenic/antiestrogenic and scavenging properties of (E)- and (Z)-resveratrol. *Life Sci* 66(9):769–777
3. Baur JA, Sinclair DA (2006) Therapeutic potential of resveratrol: the in vivo evidence. *Nat Rev Drug Discov* 5(6):493–506
4. Brinton RD (2001) Cellular and molecular mechanisms of estrogen regulation of memory function and neuroprotection against Alzheimer's disease: recent insights and remaining challenges. *Learn Mem* 8(3):121–133
5. Brookmeyer R et al (2011) National estimates of the prevalence of Alzheimer's disease in the United States. *Alzheimers Dement* 7(1):61–73
6. Bowers JL et al (2000) Resveratrol acts as a mixed agonist/antagonist for estrogen receptors alpha and beta. *Endocrinology* 141(10):3657–3667
7. Buxbaum JD et al (1992) Cholinergic agonists and interleukin 1 regulate processing and secretion of the Alzheimer beta/A4 amyloid protein precursor. *Proc Natl Acad Sci U S A* 89(21):10075–10078
8. Canistro D et al (2009) Alteration of xenobiotic metabolizing enzymes by resveratrol in liver and lung of CD1 mice. *Food Chem Toxicol* 47(2):454–461
9. Chan V et al (2011) Resveratrol improves cardiovascular function in DOCA-salt hypertensive rats. *Curr Pharm Biotechnol* 12(3):429–436
10. Collins MA et al (2009) Alcohol in moderation, cardioprotection, and neuroprotection: epidemiological considerations and mechanistic studies. *Alcohol Clin Exp Res* 33(2):206–219
11. Das DK, Maulik N (2006) Resveratrol in cardioprotection: a therapeutic promise of alternative medicine. *Mol Interv* 6(1):36–47
12. Doubek J et al. (2005) Effect of stilbene resveratrol on haematological indices of rats. *Acta Vet Brno roč. 74, č. 2, s. 205–208. ISSN 0001-7213*
13. Dubrovskaja NM et al (2009) Changes in the activity of amyloid-degrading metallopeptidases leads to disruption of memory in rats. *Zh Vyssh Nerv Dejia Im I P Pavlova* 59(5):630–638
14. El Sayed NS, Kassem LA, Heikal OA (2009) Promising therapy for Alzheimer's disease targeting angiotensin converting enzyme and the cyclooxygenase-2 isoform. *Drug Discov Ther* 3(6):307–315
15. Hashimoto S et al (2010) Expression of neutral endopeptidase activity during clinical and experimental acute lung injury. *Respir Res* 11:164
16. Hauss-Wegrzyniak B, Wenk GL (2002) Beta-amyloid deposition in the brains of rats chronically infused with thiorphan or lipopolysaccharide: the role of ascorbic acid in the vehicle. *Neurosci Lett* 322(2):75–78
17. Heo S-K et al (2008) LPS induces inflammatory responses in human aortic vascular smooth muscle cells via Toll-like receptor 4 expression and nitric oxide production. *Immunol Lett* 120(1–2):57–64
18. Howitz KT et al (2003) Small molecule activators of sirtuins extend *Saccharomyces cerevisiae* lifespan. *Nature* 425(6954):191–196

19. Huang J et al (2005) Binding of estrogen receptor beta to estrogen response element in situ is independent of estradiol and impaired by its amino terminus. *Mol Endocrinol* 19 (11):2696–2712
20. Huang T-C et al (2011) Resveratrol protects rats from A $\beta$ -induced neurotoxicity by the reduction of iNOS expression and lipid peroxidation. *PLoS One* 6(12):e29102
21. Huang J et al (2004) Estrogen regulates neprilysin activity in rat brain. *Neurosci Lett* 367 (1):85–87
22. Iwata N et al (2000) Identification of the major Abeta1-42-degrading catabolic pathway in brain parenchyma: suppression leads to biochemical and pathological deposition. *Nat Med* 6 (2):143–150
23. Iwata N et al (2002) Region-specific reduction of a beta-degrading endopeptidase, neprilysin, in mouse hippocampus upon aging. *J Neurosci Res* 70(3):493–500
24. Iwuchukwu OF, Nagar S (2008) Resveratrol (trans-resveratrol, 3,5,4'-trihydroxy-trans-stilbene) glucuronidation exhibits atypical enzyme kinetics in various protein sources. *Drug Metab Dispos* 36(2):322–330
25. Jacewicz M et al (2009) Systemic administration of lipopolysaccharide impairs glutathione redox state and object recognition in male mice. The effect of PARP-1 inhibitor. *Folia Neuropathol* 47(4):321–328
26. Jang M et al (1997) Cancer chemopreventive activity of resveratrol, a natural product derived from grapes. *Science* 275(5297):218–220
27. Juan ME et al (2005) trans-Resveratrol, a natural antioxidant from grapes, increases sperm output in healthy rats. *J Nutr* 135(4):757–760
28. Karuppagounder SS et al (2009) Dietary supplementation with resveratrol reduces plaque pathology in a transgenic model of Alzheimer's disease. *Neurochem Int* 54(2):111–118
29. Lanz T et al (1991) The role of cysteines in polyketide synthases. Site-directed mutagenesis of resveratrol and chalcone synthases, two key enzymes in different plant-specific pathways. *J Biol Chem* 266(15):99716
30. Lee VM, Goedert M, Trojanowski JQ (2001) Neurodegenerative tauopathies. *Annu Rev Neurosci* 24:1121–1159
31. Li F et al (2012) Resveratrol, a neuroprotective supplement for Alzheimer's disease. *Curr Pharm Des* 18(1):27–33
32. Lublin AL, Gandy S (2010) Amyloid-beta oligomers: possible roles as key neurotoxins in Alzheimer's disease. *Mt Sinai J Med* 77(1):43–49
33. Madani R et al (2006) Lack of neprilysin suffices to generate murine amyloid-like deposits in the brain and behavioral deficit in vivo. *J Neurosci Res* 84(8):1871–1878
34. Marambaud P, Zhao H, Davies P (2005) Resveratrol promotes clearance of Alzheimer's disease amyloid-beta peptides. *J Biol Chem* 280(45):37377–37382
35. Markus MA, Morris BJ (2008) Resveratrol in prevention and treatment of common clinical conditions of aging. *Clin Interv Aging* 3(2):331–339
36. Marr RA et al (2003) Neprilysin gene transfer reduces human amyloid pathology in transgenic mice. *J Neurosci* 23(6):1992–1996
37. McGeer EG, McGeer PL (2001) Innate immunity in Alzheimer's disease: a model for local inflammatory reactions. *Mol Interv* 1(1):22–29
38. Mokuno K et al (1994) Induction of manganese superoxide dismutase by cytokines and lipopolysaccharide in cultured mouse astrocytes. *J Neurochem* 63(2):612–616
39. Nasri S (2012) The effect of resveratrol flavonoid on learning and memory in passive avoidance and Y maze in diabetic rats. *Adv Clin Exp Med* 2012, 21(6), 705–711
40. Pervaiz S, Holme AL (2009) Resveratrol: its biologic targets and functional activity. *Antioxid Redox Signal* 11(11):2851–2897
41. Picklo MJ et al (2002) Carbonyl toxicology and Alzheimer's disease. *Toxicol Appl Pharmacol* 184(3):187–197
42. Pike CJ et al (2009) Protective actions of sex steroid hormones in Alzheimer's disease. *Front Neuroendocrinol* 30(2):239–258

43. Ruitenberg A et al (2001) Prognosis of Alzheimer's disease: the Rotterdam Study. *Neuroepidemiology* 20(3):188–195
44. Sahu SS et al. (2013) Neuroprotective effect of resveratrol against prenatal stress induced cognitive impairment and possible involvement of Atpase activity. *Pharmacol Biochem Behav* 103(3): 520–525
45. Selkoe DJ (2000) Imaging Alzheimer's amyloid. *Nat Biotechnol* 18(8):823–824
46. Shaw KN, Commins S, O'Mara SM (2001) Lipopolysaccharide causes deficits in spatial learning in the watermaze but not in BDNF expression in the rat dentate gyrus. *Behav Brain Res* 124(1):47–54
47. Sheng JG et al (2003) Lipopolysaccharide-induced-neuroinflammation increases intracellular accumulation of amyloid precursor protein and amyloid beta peptide in APP<sup>sw</sup> transgenic mice. *Neurobiol Dis* 14(1):133–145
48. Simpkins JW, Wood BE, Balin BJ (2010) Alzheimer disease: the crisis is upon us. *J Am Osteopath Assoc* 110(9 Suppl 8):Sii-2
49. Soleas GJ, Diamandis EP, Goldberg DM (1997) Resveratrol: a molecule whose time has come? And gone? *Clin Biochem* 30(2):91–113
50. Sönmez U et al (2007) Neuroprotective effects of resveratrol against traumatic brain injury in immature rats. *Neurosci Lett* 420(2):133–137
51. Sparkman NL et al (2005) Peripheral lipopolysaccharide administration impairs two-way active avoidance conditioning in C57BL/6J mice. *Physiol Behav* 85(3):278–288
52. Suzuki T, Hata S (2009) [Progresses in the researches of Alzheimer's disease to reveal the molecular mechanism of disease onset and establish the biochemical diagnostics at early stage]. *Nihon Ronen Igakkai Zasshi* 46(5):412–415
53. Swaab DF et al (2001) Structural and functional sex differences in the human hypothalamus. *Horm Behav* 40(2):93–98
54. Tang MX et al (1996) Effect of oestrogen during menopause on risk and age at onset of Alzheimer's disease. *Lancet* 348(9025):429–432
55. Vegeto E et al (2001) Estrogen prevents the lipopolysaccharide-induced inflammatory response in microglia. *J Neurosci* 21(6):1809–1818
56. Wang H-W et al (2002) Soluble oligomers of beta amyloid (1-42) inhibit long-term potentiation but not long-term depression in rat dentate gyrus. *Brain Res* 924(2):133–140
57. Williams EJ et al (2008) The effect of *Escherichia coli* lipopolysaccharide and tumour necrosis factor alpha on ovarian function. *Am J Reprod Immunol* 60(5):462–473
58. Wood JG et al (2004) Sirtuin activators mimic caloric restriction and delay ageing in metazoans. *Nature* 430(7000):686–689
59. Xiao Z-M et al (2009) Estrogen regulation of the neprilysin gene through a hormone-responsive element. *J Mol Neurosci* 39(1–2):22–26