Original Article

Neuroprotective Efficacy of *Asparagus racemosus* Root Extract on Hippocampal Neurons in Scopolamine Mouse Model of Alzheimer's Disease

Prasad Jagdish[1,*], Bhatnagar Maheep[2]

[1]Department of Zoology, Government P. G. College, Deoli, Tonk, Rajasthan, INDIA.
[2]Department of Zoology, M.L.S. University, Udaipur, Rajasthan, INDIA.

Submission Date: 15-05-2021; Revision Date: 22-07-2021; Accepted Date: 15-08-2021

Correspondence:
Dr. Prasad Jagdish,
Department of Zoology, Government P. G. College, Deoli, Tonk-304804, Rajasthan, INDIA.
Phone no: +91 9414786516

Email: jpzoology@yahoo.com

ABSTRACT

Background: Alzheimer's disease is a one of the prominent cause of senile dementia in the aged people, resulting in mental health deterioration in more than 44 millions of people worldwide. A great majority of currently available medication for neurodegenerative disorders like AD are synthetic in origin and the most of them cause severe side effects. In Ayurveda, an Indian traditional medicinal system, natural products of plant origin has used in since time immorial for treatment of neurodegenerative disorders with the minimum side effects. *Asparagus racemosus* a household plant is one of them known to possess various biological properties such as antioxidant, anti-apoptotic, adaptogen and Ayuvedic practitioners for neuronal disorders have used for it traditionally. In previous published study from this laboratory, we have shown the nootropic properties of methanolic root extract of *Asparagus racemosus* using behavioural paradigm. *Asparagus racemosus* have phytoestrogenic properties with mild estrogenic activities. Estrogen has been reported as potent neuroprotective in various *in vivo* and clinical studies. In this consequence, the aim of this study was to examine neurorprotective effects of *Asparagus racemosus* root extraction hippocampal neurons in scopolamine induced Alzheimer's type dementia in mice. **Materials and Methods:** The roots of *Asparagus racemosus* were powdered and extracted in Soxhlet apparatus. The dose of methanolic extract of AR was evaluated for its neuroprotective efficacy against scopolamine induced amnesia in mice. **Results and Conclusion:** The results of histopathological evaluation showed that methanolic root extract of *Asparagus racemosus* attenuated the neuronal death in hippocampal CA1, CA3 and DG region induced by scopolamine. These findings indicate that phytoestrogen may serve as potential preventive and therapeutic agent for neurodegenerative disorders.

Key words: Neuroprotection, *Asparagus racemosus*, Dementia, Alzheimer's disease, *in vivo*.

INTRODUCTION

Alzheimer's disease (AD) is a slowly progressive neurodegenerative disease and the most common form of dementia with memory dysfunction. AD refers to a clinical syndrome with complex pathological mechanism, resulting in memory loss and cognitive dysfunction due to progressive neurodegenration.[1] The number of person affected by dementia worldwide is estimated 46.8 million and this number reached to be near to 50 million in 2019 in which AD represents 60-80%.[2] The pathology of AD is associated with the presence of various neurotoxic protein assemblies, such as amyloid β (Aβ) plaques, development of neurofibrillary tangle (NFT_s), tau protein, alpha- synuclein, TDP-43 results in induced synaptic dysfunction and network dysfunction.[3] The behavior is an unique feature of integrated circuit with unique anatomies which related to neurotransmission and produce different behavioral phenotypes.[4] In AD, cognitive impairment, selective

SCAN QR CODE TO VIEW ONLINE

www.ajbls.com

DOI: 10.5530/ajbls.2021.10.52

loss of cholinergic neurons in hippocampus, and neuronal loss of distinct area of forebrain that results in slowly deterioration inmemory functions.[5,6] The sleep disorder is commonly reported in AD and affects lives of both the patients and the caregiver.[7] Poor sleep increases levels of the tau protein in human cerebro spinal fluid(CSF) and accelerates the spread of tau protein in neural circuits.[8] The current medications for AD treatment are symptomatic agents that purpose to improve cognitive and behavioral manifestation without altering the cause of the disease.[9] Though most of them prevent or delay the onset or slow the progression of disease.[10] Acetyl cholinesterase inhibitors, which increase the cholinergic transmission by increasing the availability of acetylcholine in cholinergic synapses[11] such as Tacrine, Rivastigmine, Donepezil are used for symptomatic treatment, but these drugs do not slow the progression of disease and are associated with various adverse effects.[12] Besides these, current treatment strategies in neurological drug development programs focus on anti-inflammatory drugs,[13] antioxidants, calcium channel blockers.[14,15] Moreover, all these drugs have limited efficacy for a limited period with their adverse side-effects.[16-18] Currently no effective and satisfactory pharmacological treatment is available for AD. Therefore, in the recent years, research concerning towards searching safe and effective is currently on medicine. In past few decades, several Ayurvedic medicine have been studied for their potential neuroprotective effects and their multi component nature.[19,20] *Asparagus racemosus* (AR), also called 'Shatavari' a native plant of the Himaliyan belt, is commonlyused traditional Indian medicine in Ayurvedic culture.[21] Shatavari described as Medhya Rasayana in Ayurveda with multi-fold benefits, specifically to improve memory and intellect. In addition, *A. racemosus* has been used for the treatment of high fever, jaundice, ulcer, inflammation and cancer. Several preclinical studies have revealed the potential implication of *A .racemosus* in the CNS disorders, such as AD, cerebral ischemia and Parkinson's disease.[22-24] *A. racemosus* root extract contains several bioactive alkaloids, phenolic compounds, flavonoids, phytosteroidal saponins (phytoestrogen-like) identified as Shatavari I –VI, sarsasapogenin, isoflavins including 8 -methoxy -5,6, 4- trihydroxyisoflavine and 7–O-β- D-glucopyroniside, aspragamine and recemosal known to possess estrogenic action.[25] Sarsasapogenin known to possess neuroprotective properties in rodent's AD model.[22] Studies have revealed the anti-parkinsonian[26] and anxiolytic activity[27] of steroidal saponins which are reported in A. racemosus. In rodents and human beings scopolamine drug used as non-selective muscarinic receptor antagonist who can produce AD like symptoms by blocking the cholinergic function of the central nervous system. Furthermore scopolamine could induce oxidative stress and amnesia by increasing AChE activity and decreasing the acetylcholine (Ach) level in the brain.[28-30] Thus, scopolamine is well-known anticholinergic drug normally used as a standard drug for inducing cognitive deficits for experimental purpose. This study aimed to investigate the neuroprotective effects of AR root extract on hippocampal neurons in scopolamine induced AD model.

MATERIALS AND METHODS

Plant material

The roots of AR were used and purchased from the local supplier. The roots were washed, shade dried, cut into small pieces, pulverized by a mechanical grinder to get a coarse powder. The powdered material was stored in airtight container for further use. The air-dried 500 gm powdered material was extracted using 100% methanol in a Soxhlet extractor for 35 hr. The solvent was evaporated at low temperature under reduced pressure in a Rota evaporator; A brownish black waxy residue completely free from the solvent was obtained. The extract gave positive tests for polyphenols, flavonoids, tannins, saponins and glycosides. The sticky concentrated mass was kept in refrigerator at 4°C and dissolved in distilled water just before use. The mice received dosages of 100 mg /kg per day based on a pilot study and reports available in the literature.

Animals

Adult Male Swiss albino mice (25 -30 mg) were used in this study. All mice were housed under controlled conditions in polyutherine cages separately for one week to acclimatize the laboratory environment. The mice were given commercial standard food pellets and water ad libitum. The mice were maintained on 12:12 hr light-dark cycle. All the experiments were performed between 0900 and 1500h. Handling and usage of animals agreed strictly with the regulation and guideline set by international norms. All protocols were made to reduce the number of animals used. The study protocol was approved by the Institutional animal ethical committee (IAEC Log.No.973/ac/06/CPCSEA).

Drugs and Chemicals

All chemicals used were purchased from commercial supplier and were of analytical grade. Scopolamine hydrobromide was purchased from Sigma –Aldrich. AR methanolic root extract was dissolved in distilled water

and administered orall. Scopolamine hydrobromide was dissolved in saline and given intraperitoneally (I.P.) after one hour of AR dose.

Methods: Experimental Design Treatment protocol

After acclimatization for one week, the mice were randomly distributed into four different groups with six mice in each group and were maintained under similar condition during the experiment. Scopolamine (1 mg/kg b.w.) was dissolved in saline and givensingle dose/ day intraperitoneally for 7 days. All the drugs were administered in the morning session i.e. 8.00-9.00 AM each day.

Histopathology After completion of treatment protocol mice were anesthetized with Euthanal and sacrificed by decapitation on next day. Their whole brains were removed from the skull and fixed in 10% neutral chilled formalin for 18 hr at 4°C. The brains were dehydrated in graded ethanol series. They were cleared in xylene and immersed in Xylene 50% + paraffin wax 50 % mixture at 60°C. Tissue was then transferred to 100% paraffin wax at 58°C. Brains were embedded in paraffin wax and left at 4°C until used. The paraffin blocks were serially sectioned at 10µm thickness on a rotatory microtome. Coronal sections containing hippocampal sub region (bregma -1.46 mm to -2.18 mm) were used for histological study. Sections were dewaxed in xylene and then passed through descending series of ethanol and finally hydrated. After that, sections were stained in 1% cresyl violet according routine laboratory procedures, dehydrated in ascending graded alcohol series, cleared in xylene. Sections were mounted in Distren Plastcizer Xylene (DPX). Qualitative analysis of neurons in the pyramidal cell layer of Ammon's horn and granular layer of DG of hippocampus was performed, microphotographs of each section was taken using Olympus BX 51 microscope. The cells with significant granules were counted. The images were analysed using OLYSIA Autobioreport software. Medium or large neurons with distinct nucleus and intact cell membrane were considered as viable. Darkly stained pyknotic neurons with diffused or fragmented nuclei were considered as apoptotic or degenerated.

Statistical Analysis

All the results were expressed as mean± SEM (Standard error of mean). All results and data were statically analysed using one-way ANOVA, $p<0.05$ was considered to be statically significant.

Table 1: Animal groups and treatment protocol.

Group		Treatment and Duration
Group I *N*=6	Control	Mice were received vehicle for a period of one week.
Group II *N*=6	AR	Mice were administrated a single dose of AR methanolic roots extract (100 mg/kg b.w. P.O) for a period of one week.
Group III *N*=6	SCO	Mice were received a single dose of Scopolamine (1mg/kg b.w. I.P.) for a period of one week.
Group IV *N*=6	SCO +AR	Mice were administrated AR methanolic roots extract (100 mg/kg P.O.) + Scopolamine (1mg/kg I.P.) for a period of one week.

(All protocol and research designed under Ph.D. supervisor)

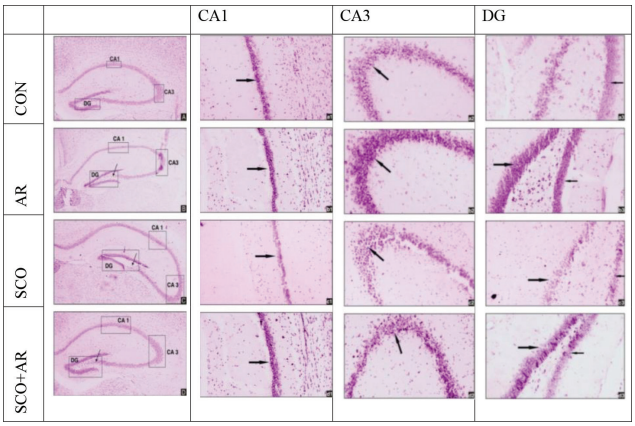

Figure 1: Effects of *Asparagus racemosus* methanolic root extract on the neuron cell bodies in hippocampus in scopolamine induced amnesia in mice.

RESULTS

Qualitative analysis of neuron cell bodies

Histopathological changes were observed in mice hippocampus (Figure 1A-D). The scopolamine treated group showed extensive injuries of pyramidal cells in hippocampal sub regions CA1, CA3 and DG (Figure 1C). In this consequence, the effects of AR methanolic root extract in the hippocampus of mice treated with scopolamine were observed. In scopolamine treated group cells appeared to have vacuolation, hyper chromatic, pyknotic, busted morphology without distinct cell membrane chromatolysis appeared in some cells (Figure 1 c1-c3). Scopolamine also produces selective staining of nissil granules, glial activation, presence of dendritic cytoplasm suggesting protein denaturing and neurodegeneration in the hippocampus and dentate gyrus neuron cell bodies. In mice, treated with AR root extract combined with scopolamine

(Figure 1D), showed a marked reappearance of CA pyramidal cell layers as well as reduction of neuronal loss and neurodegeneration in all the three regions studied CA1, CA3 and DG (Figure 1 d1-d3).

Note changes in cell bodies of CA1, CA3 and DG regions (Figure A-D Marked area). Normal cytoarchetecture in a hippocampus formation of control group (Figure A) and *A. racemosus* group (Figure B). Detail of all three region CA1, CA3 and DG in control group (Arrow Figure a1, a2 and a3) and *A.racemosus* group (Arrow Figure b1, b2 and b3). Hippocampus formation of mouse of scopolamine group showing high neuronal damage, neuronal loss and neurodegeneration in CA1, CA3 and DG (Figure C). Detail of all three region in scopolamine group (Arrow Figure c1, c2 and c3). In mice, treated with *A. racemosus* root extract combined with scopolamine (Figure D), showing a marked reappearance of CA pyramidal cell layers as well as reduction of neuronal loss and neurodegeneration in all the three regions studied CA1, CA3 and DG (Detail of all three region Figure d1, d2 and d3).

Quantitative analysis of neuron cell bodies

The number of viable neurons of CA1 and CA3 sub-region of hippocampus showed significant decline in scopolamine group (SCO: CA1: 6.240 ±0.190 CA3: 6.953 ±0.439 $p<0.005$) as compared to control (Control: CA1: 7.131 ±1.396 CA3: 11.127 ±0.917 $p<0.005$) (Figure 2A, 2B,). Simultaneous treatment of AR and scopolamine in mice caused a significant elevation in the number of viable neurons of CA1 and CA3 sub-region of hippocampus (AR+SCO: CA1: 8.285 ±1.16 CA3: 8.954 ±0.463 $p<0.005$) as compared to those treated with scopolamine alone (SCO: CA1: 6.240 ±0.190 CA3: 6.953 ±0.439 $p<0.005$) (Figure 2A, 2B,). However, no significant difference in the of viable neurons of CA1 and CA3 sub-region of hippocampus was observed between control (Control: CA1: 7.131 ±1.396 CA3: 11.127 ±0.917 $p<0.005$) and AR (AR: CA1: 7.989 ±1.55 CA3: 8.989 ±1.55 $p<0.005$) treated groups (Figure 2A, 2B,). The number of viable neurons of DG region of hippocampus showed significant decline in scopolamine group (SCO: DG: 15.206 ±1.791 $p<0.005$) as compared to control (CONTROL: DG: 19.317 ±1.294 $p<0.005$) (Figure 2C) Simultaneous treatment of AR and scopolamine in mice caused a significant elevation in the number of viable neurons of DG region of hippocampus (AR+SCO: DG: 17.141 ±1.252 $p<0.005$) as compared to those treated with scopolamine alone (SCO: DG: 15.206 ±1.791 $p<0.005$) However, no significant difference in the of viable neurons of DG region of hippocampus was observed between control

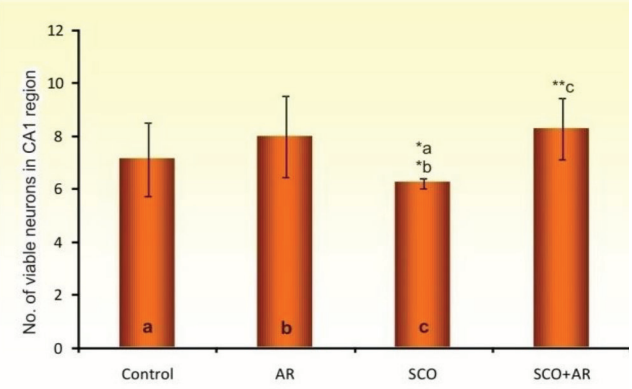

Figure 2A: Viable neuron count in CA1 region Mean ±SE (*P*<0.001),*(*P*<0.005).**

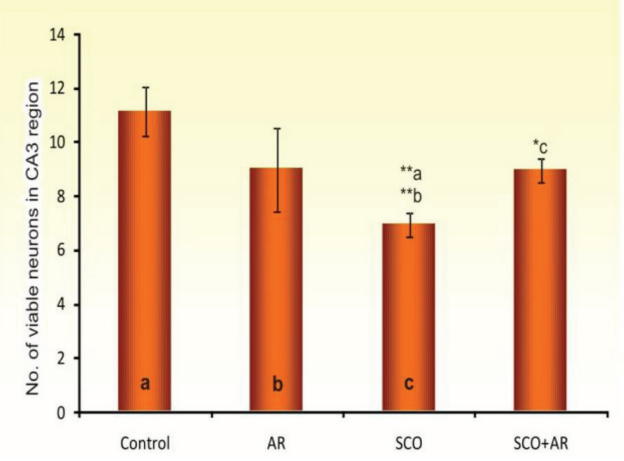

Figure 2B: Viable neuron count in CA3region mean±SE (*P*<0.005),* (*P*<0.001).**

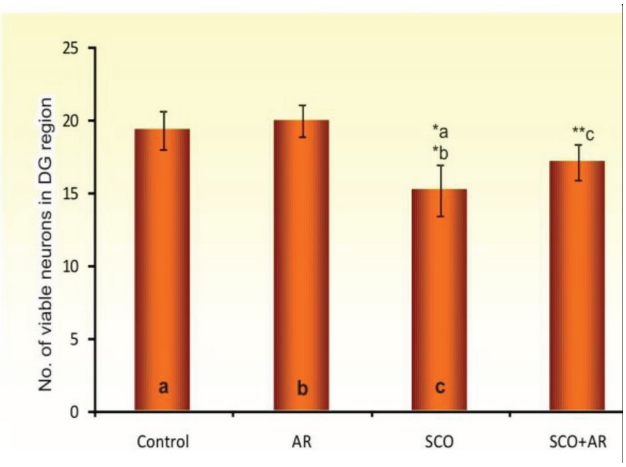

Figure 2C: Viable neuron count in DG region mean ± SE (*P*<0.001), * (*P*<0.005).**

Figure 2: Effects of *Asparagus racemosus* methanolic root extract on the viable neuron count in hippocampus in scopolamine induced amnesia in mice (Quantitative assessment of viable neuron).

(Control: DG: 19.317 ±1.294 p<0.005) and AR (AR: DG: 19.972 ±1.074p<0.005) treated groups (Figure 2C). Cell count in scopolamine group showing significant decrease in viable neuron number in CA 1, CA3 and DG sub region of hippocampus of mice as compared to control and AR group (Figure 2A, 2B, 2C).In mice, treated with *A. racemosus* root extract combined with scopolamine, showing a significant increase in cell number in three regions studied CA1, CA3 and DG (Figure 2A, 2B, 2C).

DISCUSSION

Alzheimer's disease is considered the most common type dementia with progressive decline both in memory and cognitive ability.[31] Histopathogy of AD brain is characterized by neurodegeration in the DG of hippocampus, temporal cortex, deposition of neurofibrillary tangles and senile plaque.[32,33] Worldwide, approximately 44 million people are suffering with AD or related form of dementia.[34] Experimental and clinical studies have indicated that estrogen and estrogen like molecule widely influence brain functions by acting as antioxidant,[35] supports DNA repair,[36]synthesis of growth factors,[37] synaptic plasticity,[38]and the blood flow to the brain,[39] It has been reported that estrogen modulate cell proliferation,[40] cell differentiation,[41] cell survival,[42] and also delay the onset of AD by modulating cholinergic functions.[43]Recent studies suggested that HRT with estrogen for post-menopausal women is effective in preventing AD and PD.[44] Series of clinical and laboratory evidences highlighted the medicinal importance of herbal preparation in the management of neuronal disorders such as depression,[45] anxiety,[46,47] epilepsy,[48,49] and also inneurodegenerative diseases.[50] Phytoestrogens are plant originated estrogens, have received increased investigation due to their estrogen like activities and competing the estrogen hormone at receptors bindings. They are capable to enhance learning, cognitive ability, promote differentiation in various type of stem cell including neural stem/progenitor cells (NSPCs).[51,52] Several clinical and laboratory studies have revealed that phytoestrogens treatment could improve cognitive decline in post-menopausal women and *in vivo* rodent models.[53] *Asparagus racemosus* (AR) is well-documented for its potent phytoestrogenic properties. In Ayurveda AR is described as rasayana, cited as nervine tonic, and also accepted as female tonic due to its phytoestrogenic properties.[21] Saxena *et al.,* 2010; Panday *et al*., 2005[54,55] have shown that roots and leaves of AR possess estrogenic properties.Sarsasapogenin, a steroid saponinpresent in AR is recently identified as anti-amyloidogenic.[56] Kasyap *et al*., 2020[56] have shown that AR based pharmaceutical preparation demonstrat anti-neurodegenerative properties against oxidative stress-induced neurotoxicity in PC12 cells. Lalert *et al*., 2018[57] have demonstrated thatAR promotes the release of neurotrophic factor BDNF in the prefrontal cortex and hippocampus of the brain. Phytoestrogens such as sarsaponin, protodioscin and diosgeninact as a precursor of progesteron and has been also reported in AR roots.[58-60] Three classes of phytoestrogens, flavonoids, isoflavones and coumestans extracted from AR, possess the most potent estrogenic activity.[61] Alzheimer's disease and neurodegenrative disease are less common in women before menopause. It's possible that estrogen protect from neurodegeneration. But this protection lost with gradual decline in estrogen level.[62,63] Studies have also established estrogen as strong neuroprotactant and neuroplastic hormone within CNS.[64,65] In animal model estrogen deficiency increased the vulnerability in neuronal elements such as cholinergic neurons of basal forebrain and hippocampus, perhaps estrogen played an important role in improving synaptic plasticity, preventing neuronal loss by stimulating axonal and synaptic regeneration and decreasing inflammation,[66,67] reducing tau protein and accelerate break down of APP to amyloid beta protein (AB).[68] Scopolamine induced amnesic rodent model is one of the well-established animal model for studying memory dysfunctions,[69,70] Scopolamine, a non-selective muscarinic antagonist block cholinergic neurotransmission and induces the deficit in the learning, acquisition and memory processing.[71,72] Both the long term and short term memory deficit are produced by scopolamine.[73,74] Similarities have been observed between patient with Alzheimer's disease and scopolamine treated animals. Thus scopolamine can provide a use full pharmacological tool to generate a partial model of the disorder such as AD.[75] The herbal remedies containing *Asparagus racemosus* with *Withania somenifera* and *Emblica officinalis* was demonstrated antistress activity in rat.[76] Parihar and himnani 2004[77] was evaluated the potential of methanolic root extract of *Asparagus racemosus* against kainic acid induced hippocampus and striatal neurodegeneration in mice. The cerebroprotective effect of *Asparagus* rhizome in cerebral ischemia induced by bilateral carotid artery occlusion in rats also studied.[78] This study demonstrated the neuroprotective role of methanolic root extract of *Asparagus racemosus* on scopolamine induced memory impairment association with protecting the neuronal loss of hippocampus. Laddawan *et al.* 2013[79] reported that *Asparagus racemosus*

root extract enhance neuronal cell viability in the CA1 and CA3 sub region of hippocampus and medial prefrontal cortex which are implicated in recognition memory. It has been reported that *Asparagus racemosus* shows Phytoestrogenic properties due to its steroidal saponins namely shatavarins.[80] These saponins increased memory and protected scopolamine induce amnesia in rodents.[81] In the present study we observed similar Phytoestrogenic effects of methanolic root extract of *Asparagus racemosus*.

In this study, comparison of Histopathological observations of changes in hippocampal regions CA1 to CA4 and DG in control with scopolamine treated group showed extensive cell injuries in CA1, CA3 and DG demonstrating vacuolation, hyperchromatic, pyknotic, busted morphology without distinct cell membrane chromatolysis appeared in some cells. Selective loss of nissil staining, glial activation and presence of dendritic cytoplasm suggest protein denaturation and neurodegeneration in these cell bodies. In mice,treated with AR root extract combined with scopolamine, showed marked changes cellular appearance of all the three layers. A significant number of normal cells were observed throughout all the three layers. Quantitative studies of cells in these regions supported the histopathological observations.

The number of viable neurons of CA1 and CA3 sub-region of hippocampus showed significant decline in CA1, CA3 and DG of scopolamine treated mice hippocampus as compared to control. Combined treatment with AR and scopolamine caused a significant increase in the number of viable neurons in CA1 and CA3 sub-region of hippocampus as compared to those treated with scopolamine alone. No significant change in the viable neurons of CA1 and CA3 sub-region of hippocampus was observed between control and only AR treated brain. Quantitative studies of scopolamine treated DG region showed significant decline as compared to control. While AR and scopolamine brain showed significant elevation in the number of viable neurons as compared to those treated with scopolamine alone. However, no significant difference in the DG region was observed between control and AR treated brain. Observation of significantly reduced neurodegenerative changes in neuron cell bodies hippocampal layers (CA1, CA3 and DG) in present investigation after treatment with AR methanolic root extract (100 mg/ kg b.w.) demonstrate that AR extract is a potential candidate for developing safe and effective alternative therapy.

CONCLUSION

The present study concludes that the methanolic root extract (100 mg/ kg b.w.) of AR produce neuroprotective effects against the scopolamine induced amnesia. Based on qualitative and quantitative observations in hippocampal neuron cell bodies we suggest that that AR methanolic root extract may be a promising candidate for developing effective the treatment for AD like neurodegenrative disorders.

ACKNOWLEDGEMENT

This work is highly acknowledged to UGC CRO Bhopal and MLSU Udaipur for providing necessary facilities for the research.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

AR: *Asparagus racemosus*; **AD:** Alzheimer's disease; **CA:** Cornu Ammonis; **DG:** Dentate gyrus; **CNS:** Central Nervous System; **DNA:** Deoxyribose Nucleic Acid; **DPX:** Ditrene Plasticizer Xylene; **mAChRs:** Muscarinic ACh Receptors; **mg:** Milli Grams; **ppm:** Parts Per Million; **SVZ:** Subventricular-zone; **SGZ:** Subgranular-zone; **TDP43:** TAR DNA-binding protein 43; **NFT:** Neurofibrillary tangles; **CSF:** Cerebrospinal fluid; **PD:** Parkinson's disease; **AchE:** Acetyl cholinesterase; **Ach:** Acetylcholine; **SEM:** Standard error of mean; **ANOVA:** Analysis of variance.

SUMMARY

The population ageing has major social and economic consequences and poses a significant threat to global prosperity. Older people are affected by Alzheimer's and Parkinson's disease and mental health problems. Schizophrenia, depression, postmenstrual syndrome, postnatal and depression are abundant in ageing population. Dementia is a disease conditions in which there is decline in brain function, cognition, behavior, and other thinking skills that affects a person's ability to perform everyday activities. This condition occurs due to neuronal in the brain. These neurodegenerative diseases are associated with gradual loss of memory and impaired cognitive functions. Prevalence of neuronal diseases is greater in women than men after the age of 65 or in their post-menopausal stage. The level of estrogens rapidly decline in women in their post-menopausal stage and makes them more vulnerable to

neurodegenerative diseases. Estrogen is capable to delay the onset of Alzheimer's disease (AD) by modulating activity of cholinergic neurons, metabolism of monoamine and BDNF mRNA essential for optimal brain function. Estrogen strongly influences memory and cognition as well as brain functions. Phytoestrogens, which are plant derived, xenoestrogens are found in seeds, fruits, and vegetables. In present study, we used scopolamine induced amnesic mice model to study the neuroprotective and neuromodulatory effect of *Asparagus racemosus:* as potent phytoestrogen. *Asparagus racemosus* (Family: Aparagaceae), called Shatavari plant is described as queen of herbs in the ayurvedic system of medicine for its multidimensional therapeutic values. In the present study attempts have been made to evaluate effects of methanolic root extract of *Asparagus racemosus* on learning and memory in scopolamine induced dementic mice model of Alzheimer's disease. Scopolamine induce a wide range of neurodegenrative changes in brain and causes neurochiatric disorders. Histological evidences in present study have demonstrated hippocampal neurodegeneration in scopolamine treated mice. *Asparagus racemosus* treatment resulted in significant neuroprotection against scopolamine induced neurotoxicity. Methanolic root extract of *Asparagus racemosus* effectively reversed the neurodegenerative changes in CA1, CA3 and DG sub regions of hippocampus. Further significant ameliorative effect against neurodegeneration proves neuroprotective activity of phytoestrogens of *Asparagus racemosus*. Qualitative and quantitative study was also carried to demonstrate protective effect of Shatavari root. Differential counting of degenerating cells (dark cells, apoptotic neuronal cell bodies, chromatolytic neurons, vacuolated cytoplasm) in scopolamine treated groups have shown significant changes in cell number in CA1 CA3 and DG sub field of hippocampus. Maximum normal cell count in these sub regions was observed in *Asparagus racemosus* treated mice as compared to scopolamine treated mice.

REFERENCES

1. Uddin MS, Mamun AA, Hossain MS, Ashaduzzaman M, Noor MAA, Hossain MS, Uddin MJ, Sarker J, Asaduzzaman MNeuroprotective Effect of <i>Phyllanthus acidus</i> L. on Learning and Memory Impairment in Scopolamine-Induced Animal Model of Dementia and Oxidative Stress: Natural Wonder for Regulating the Development and Progression of Alzheimer's Disease. Adv Alzheimers Dis. 2016;05(2):53-72. doi: 10.4236/aad.2016.52005.
2. Alzheimer's Association. Alzheimer's disease facts and figures. Alzheimers Dem. 2020;15:5-25.
3. Mucke L, Selkoe DJ. Neurotoxicity of amyloid beta-protein: synaptic and network dysfunction. Cold Spring HarbPerspectMed. 2012;2(7):a006338. doi: 10.1101/cshperspect.a006338, PMID 22762015.
4. Siddiqi SH, Taylor SF, Cooke D, Pascual-Leone A, George MS, Fox MD. Distinct symptom-specific treatment targets for circuit-based neuromodulation. AmJPsychiatry. 2020;177(5):435-46. doi: 10.1176/appi.ajp.2019.19090915, PMID 32160765.
5. Adefegha SA, Oboh G, Olasehinde TA. Alkaloid extracts from shea butter and breadfruit as potential inhibitors of monoamine oxidase, cholinesterases, and lipid peroxidation in rats' brain homogenates: A comparative study. CompClinPathol. 2016;25(6):1213-9. doi: 10.1007/s00580-016-2331-0.
6. Oboh G, Adewuni TM, Ademosun AO, Olasehinde TA. Sorghum stem extract modulates Na+/K+-ATPase, ecto-5′-nucleotidase, and acetylcholinesterase activities. CompClinPathol. 2016;25(4):749-56. doi: 10.1007/s00580-016-2259-4.
7. Borges CR, Poyares D, Piovezan R, Nitrini R, Brucki S. Alzheimer's disease and sleep disturbances: a review. Arq Neuropsiquiatr. 2019;77(11):815-24. doi: 10.1590/0004-282X20190149, PMID 31826138.
8. Wang C, Holtzman DM. Bidirectional relationship between sleep and Alzheimer's disease: role of amyloid, tau, and other factors. Neuropsychopharmacology. 2020;45(1):104-20. doi: 10.1038/s41386-019-0478-5, PMID 31408876.
9. Atri A. The Alzheimer's disease clinical spectrum: Diagnosis and management. Med Clin North Am. 2019;103(2):263-93. doi: 10.1016/j.mcna.2018.10.009, PMID 30704681.
10. Cummings J, Lee G, Ritter A, Sabbagh M, Zhong K. Alzheimer's disease drug development pipeline: 2020. Alzheimers Dement (N Y). 2020;6(1):e12050. doi: 10.1002/trc2.12050. PMID 32695874.
11. Giacobini E. Cholinesterase inhibitors: from the Calabar Bean to Alzheimer therapy. Cholinesterase and Cholinesterase Inhibitors 2000:181-226.
12. Knapp MJ. A 30-week randomized controlled trial of high-dose tacrine in patients with Alzheimer's disease. The Tacrine Study Group. JAmMedAssoc. 1994;271(13):985-91. doi: 10.1001/jama.271.13.985.
13. McGeer PL, McGeer EG. NSAIDs and Alzheimer disease: epidemiological, animal model and clinical studies. NeurobiolAging. 2007;28(5):639-47. doi: 10.1016/j.neurobiolaging.2006.03.013, PMID 16697488.
14. Rinaldi P, PolidoriMC, Metastasio A, Mariani E, Mattioli P, Cherubini A, Catani M, Cecchetti R, Senin U, Mecocci P. Plasma antioxidants are similarly depleted in mild cognitive impairment and in Alzheimer's disease. NeurobiolAging. 2003;24(7):915-9. doi: 10.1016/s0197-4580(03)00031-9, PMID 12928050.
15. Yasar S, Corrada M, Brookmeyer R, Kawas C. Calcium channel blockers and risk of AD: theBaltimore Longitudinal Study of Aging. NeurobiolAging. 2005;26(2):157-63. doi: 10.1016/j.neurobiolaging.2004.03.009, PMID 15582745.
16. Zhou X, Li Y, Shi X, Ma C. An overview on therapeutics attenuating amyloid beta level in Alzheimer's disease: targeting neurotransmission, inflammation, oxidative stress and enhanced cholesterol levels. AmJ TranslRes. 2016;8(2):246-69. PMID 27158324.
17. Lu C, Dong L, Lv J, Wang Y, Fan B, Wang F, Liu X. (S)-protopanaxadiol (PPD) alleviates scopolamine-induced memory impairment via regulation of cholinergic and antioxidant systems, and expression of Egr-1, c-Fos and c-Jun in mice. ChemBiolInteract. 2018;279:64-72. doi: 10.1016/j.cbi.2017.11.008, PMID 29133030.
18. Jeon SJ, Kim B, Park HJ, Zhang J, Kwon Y, Kim DH, Ryu JH. The ameliorating effect of 1-palmitoyl-2-linoleoyl-3-acetylglycerol on scopolamine-induced memory impairment via acetylcholinesterase inhibition and LTP activation. BehavBrainRes. 2017;324:58-65. doi: 10.1016/j.bbr.2017.01.040, PMID 28137622.
19. Balkrishna A, Misra L. Chemo-botanical and neurological accounts of some ayurvedic plants useful in mental health. NatProdJ. 2018;8(1):14-31. doi: 10.2174/2210315507666170616082903.
20. Narayanan G, Prabhu K, Chaudhuri A, Rao MRK, Selvi VSK, Balaji TK, Kanagasabai V, Dinakar S. Neuro-protective effect of Ayurveda formulation, Saraswatharishtam, on scopolamine induced memory impairment in animal model. PharmacognJ. 2020;12(3):465-72. doi: 10.5530/pj.2020.12.72.
21. GoyalRK, Singh J, Lal H. Asparagus racemosus--an update. Indian J MedSci.2003;57(9):408-14. PMID 14515032.
22. Pan H, Van Khang P, Dong D, Wang R, Ma L. Synthesis and SAR study of novel sarsasapogenin derivatives as potent neuroprotective agents and NO production inhibitors. Bioorg Med Chem Lett. 2017;27(3):662-5. PMID: 27919659. Doi: 10.1016/j.bmcl.2016.11.070

23. Bagchi P, Anuradha M, Kar A. Ayur-Informtics: establishing an Ayurvedic medication for Parkinson's disorder. International Journal of Advances in Chemical Engineering and Biological Science.2017;4:21-5.
24. Nandagopal M, Muralidhar P, Thirumurug G. Cerebroprotective Effect of Root Extract of Asparagus racemosus Willd. in Global Cerebral Ischemia in Rats. J Pharmacol Toxicol. 2010;6(1):49-61. doi: 10.3923/jpt.2011.49.61.
25. Kumeta Y, Maruyama T, Wakana D, Kamakura H,Goda Y. Chemical analysis reveals the botanical origin of shatavari products and confirms the absence of alkaloid asparagamine A in *Asparagus racemosus*. J Na tMed. 2013;67(1):168-73. doi: 10.1007/s11418-012-0669-4, PMID 22529051.
26. Smita SS, Raj Sammi SS, Laxman TS, Bhatta RS, Pandey R. Shatavarin IV elicits lifespan extension and alleviates Parkinsonism in Caenorhabditis elegans. Free RadicRes. 2017;51(11-12):954-69. doi: 10.1080/10715762.2017.1395419, PMID 29069955.
27. Garabadu D, Krishnamurthy S. *Asparagus racemosus* attenuates anxiety-like behavior in experimental animal models. CellMolNeurobiol. 2014;34(4):511-21. doi: 10.1007/s10571-014-0035-z, PMID 24557501.
28. Balaban H, Nazıroğlu M, Demirci K, Övey İS. The protective role of selenium on scopolamine-induced memory impairment, oxidative stress, and apoptosis in aged rats: The involvement of TRPM2 and TRPV1 channels. MolNeurobiol. 2017;54(4):2852-68. doi: 10.1007/s12035-016-9835-0, PMID 27021021.
29. Han SH, Kim SJ, Yun YW, Nam SY, Lee HJ, Lee BJ. Protective effects of cultured and fermented ginseng extracts against scopolamine-induced memory loss in a mouse model. LabAnimRes. 2018;34(1):37-43. doi: 10.5625/lar.2018.34.1.37, PMID 29628975.
30. Lee HL, Lim SA, Lee HW, Yoo HR, Kim HG. Yuk-Mi-Jihwang-Tang, a traditional Korean multiple herbal formulae, improves hippocampal memory on scopolamine injection-induced amnesia model of C57BL/6 mice. EvidBased ComplementAlternatMed. 2018Jan1;2018:2821040. doi: 10.1155/2018/2821040, PMID 29507588.
31. Alzheimer's Association. Alzheimer's disease facts and figures. Alzheimers Dem. 2018;14(3):367-429. doi: 10.1016/j.jalz.2018.02.001.
32. Al Mamun A, Uddin MS, Kabir MT, Khanum S, Sarwar MS, Mathew B, Rauf A, Ahmed M, Ashraf GM. Exploring the promise of targeting ubiquitin-proteasome system to combat Alzheimer's disease. NeurotoxRes. 2020;38(1):8-17. doi: 10.1007/s12640-020-00185-1, PMID 32157628.
33. Rahman MA, Rahman MR, Zaman T, UddinMS, Islam R, Abdel-Daim MM, Rhim H. Emerging potential of naturally occurring autophagy modulators against neurodegeneration. CurrPharmDes. 2020;26(7):772-9. doi: 10.2174/1381612826666200107142541, PMID 31914904.
34. Uddin MS, Kabir MT, Niaz K, Jeandet P, Clément C, Mathew B, *et al*. Molecular insight into the therapeutic promise of flavonoids against Alzheimer's disease. Molecules. 2020;25(6):1267. doi: 10.3390/molecules25061267, PMID 32168835.
35. Kumar S, Lata K, Mukhopadhyay S, Mukherjee TK. Role of estrogen receptors in pro-oxidative and anti-oxidative actions of estrogens: A perspective. BiochimBiophys Acta. 2010;1800(10):1127-35. doi: 10.1016/j.bbagen.2010.04.011, PMID 20434525.
36. Bethea CL, Kohama SG, Reddy AP, Urbanski HF. Ovarian steroids regulate gene expression in the dorsal raphe of old female macaques. NeurobiolAging. 2016;37:179-91. doi: 10.1016/j.neurobiolaging.2015.10.004, PMID 26686671.
37. Scharfman HE, MacLusky NJ. Estrogen and brain-derived neurotrophic factor (BDNF) in hippocampus: Complexity of steroid hormone-growth factor interactions in the adult CNS. FrontNeuroendocrinol. 2006;27(4):415-35. doi: 10.1016/j.yfrne.2006.09.004, PMID 17055560.
38. McEwen BS, Milner TA. Hippocampal formation: shedding light on the influence of sex and stress on the brain. Brain ResRev. 2007;55(2):343-55. doi: 10.1016/j.brainresrev.2007.02.006, PMID 17395265.
39. Chambliss KL, Yuhanna IS, Mineo C, Liu P, German Z, Sherman TS, Mendelsohn ME, Anderson RG, Shaul PW. Estrogen receptor alpha and endothelial nitric oxide synthase are organized into a functional signaling module in caveolae. CircRes. 2000;87(11):E44-52. doi: 10.1161/01.res.87.11.e44, PMID 11090554.
40. Molina L, Bustamante F, Ortloff A, Ramos I, Ehrenfeld P, Figueroa CD. Continuous exposure of breast cancer cells to tamoxifen upregulates GPER-1 and increases cell proliferation. FrontEndocrinol. 2020;11:563165. doi: 10.3389/fendo.2020.563165.
41. Imanishi T, Kobayashi K, HanoT, Nishio I. Effect of estrogen on differentiation and senescence in endothelial progenitor cells derived from bone marrow in spontaneously hypertensive rats. HypertensRes. 2005;28(9):763-72. doi: 10.1291/hypres.28.763, PMID 16419650.
42. Yu X, Zhang X, Dhakal IB, Beggs M, Kadlubar S, Luo D. Induction of cell proliferation and survival genes by estradiol-repressed microRNAs in breast cancer cells. BMC Cancee 2012;12(1):1-13. doi: 10.1186/1471-2407-12-29, PMID 22260523.
43. Mateos L, Persson T, Katoozi S, Gil-Bea FJ, Cedazo-Minguez A. Estrogen protects against amyloid-beta toxicity by estrogen receptor alpha-mediated inhibition of Daxx translocation. NeurosciLett. 2012;506(2):245-50. doi: 10.1016/j.neulet.2011.11.016, PMID 22119000.
44. Song YJ, Li SR, Li XW, Chen X, Wei ZX, Liu QS, Cheng Y. The Effect of estrogen Replacement Therapy on Alzheimer's disease and Parkinson's disease in Postmenopausal Women: A Meta-Analysis. FrontNeurosci. 2020;14:157. doi: 10.3389/fnins.2020.00157, PMID 32210745.
45. Akhondzadeh S, Fallah-Pour H, Afkham K, Jamshidi AH, Khalighi-Cigaroudi F. Comparison of Crocus sativusL. and imipramine in the treatment of mild to moderate depression: a pilot double-blind randomized trial. BMC ComplementAlternMed. 2004;4:12. doi: 10.1186/1472-6882-4-12, PMID 15341662.
46. Movafegh A, Alizadeh R, Hajimohamadi F, Esfehani F, Nejatfar M. Preoperative oral Passiflora incarnata reduces anxiety in ambulatory surgery patients: A double-blind, placebo-controlled study. AnesthAnalg. 2008;106(6):1728-32. doi: 10.1213/ane.0b013e318172c3f9, PMID 18499602.
47. Woelk H, ArnoldtKH, Kieser M, Hoerr R. Ginkgo biloba special extract EGb 761 in generalized anxiety disorder and adjustment disorder with anxious mood: a randomized, double-blind, placebo-controlled trial. JPsychiatrRes. 2007;41(6):472-80. doi: 10.1016/j.jpsychires.2006.05.004, PMID 16808927.
48. Buenafe OE, Orellana-Paucar A, Maes J, Huang H, Ying X, De Borggraeve W, Crawford AD, Luyten W, Esguerra CV, de Witte P. Tanshinone IIA exhibits anticonvulsant activity in zebrafish and mouse seizure models. ACS ChemNeurosci. 2013;4(11):1479-87. doi: 10.1021/cn400140e, PMID 23937066.
49. Rahmati B, Khalili M, Roghani M, Ahghari P. Anti-epileptogenic and antioxidant effect of Lavandula officinalis aerial part extract against pentylenetetrazol-induced kindling in male mice. JEthnopharmacol. 2013;148(1):152-7. doi: 10.1016/j.jep.2013.04.004, PMID 23603193.
50. Bhatnagar M, Shukla SD, Bhatnagar R. Experimental neurodegeneration in hippocampus and its phytoremidation. JHerbPharmacother. 2005;5(2):21-30. doi: 10.1080/J157v05n02_03, PMID 16260407.
51. Heim M, Frank O, Kampmann G, Sochocky N, Pennimpede T, Fuchs P, Hunziker W, Weber P, Martin I, Bendik I. The phytoestrogen genistein enhances osteogenesis and represses adipogenic differentiation of human primary bone marrow stromal cells. Endocrinology. 2004;145(2):848-59. doi: 10.1210/en.2003-1014, PMID 14605006.
52. Oberbauer E, Urmann C, Steffenhagen C, Bieler L, Brunner D, Furtner T, Humpel C, Bäumer B, Bandtlow C, Couillard-Despres S, Rivera FJ, Riepl H, Aigner L. Chroman-like cyclic prenylflavonoids promote neuronal differentiation and neurite outgrowth and are neuroprotective. JNutrBiochem. 2013;24(11):1953-62. doi: 10.1016/j.jnutbio.2013.06.005, PMID 24070601.
53. Pan M, Li Z, Yeung V, Xu RJ. Dietary supplementation of soy germ phytoestrogens or estradiol improves spatial memory performance andincreases gene expression of BDNF, TrkB receptor and synaptic factors in ovariectomized rats. NutrMetab (Lond). 2010;7(1):75. doi: 10.1186/1743-7075-7-75, PMID 20843342.
54. Saxena VK, Chourasia S. A new isoflavone from the roots of *Asparagus racemosus*. Fitoterapia. 2001Mar1;72(3):307-9. doi: 10.1016/s0367-326x(00)00315-4, PMID 11295314.
55. Pandey SK, Sahay A, Pandey RS, Tripathi YB. Effect of Asparagus racemosus rhizome (Shatavari) on mammary gland and genital organs of pregnant rat. Phytother Res. 2005;19(8):721-4. doi: 10.1002/ptr.1590. PMID 16177978.
56. Kashyap P, Muthusamy K, Niranjan M, Trikha S, Kumar S. Sarsasapogenin: A steroidal saponin from *Asparagus racemosus* as multi target directed ligand in Alzheimer's disease. Steroids. 2020 Jan;153:108529. doi: 10.1016/j.steroids.2019.108529. Epub 2019 Oct 28. PMID: 31672628.

57. Lalert L, Kruevaisayawan H, Amatyakul P, IngkaninanK, Khongsombat O. Neuroprotective effect of *Asparagus racemosus* root extract via the enhancement of brain-derived neurotrophic factor and estrogen receptor in ovariectomized rats. JEthnopharmacol. 2018;225:336-41. doi: 10.1016/j.jep.2018.07.014, PMID 30009979.
58. Shao Y, Poobrasert O, Kennelly EJ, Chin CK, Ho CT, Huang MT, Garrison SA, Cordell GA. Steroidal saponins from Asparagus officinalis and their cytotoxic activity. Planta Med. 1997;63(3):258-62. doi: 10.1055/s-2006-957667, PMID 9225609.
59. Modaresi M, Mahdian B. The effect of hydroalcoholic extract of Trigonella foenum- Graceum L. on reproductive system in Balb/c. JHerb Drugs. 2012;2:261-7.
60. Mokhtari M, Shariatie M, Ghahramanie R. Effect of Trigonella foenum-Graceum L.Seed extract on concentration of testosterone and spermatogenesis in rats. JMed Plants. 2008;7(25):12-20.
61. Murkies AL, Wilcox G, Davis SR. Phytoestrogens. JClinEndocrinolMetab. 1998;83(2):297-303. doi: 10.1210/jcem.83.2.4577, PMID 9467531.
62. Zhao L, Woody SK, Chhibber A. Estrogen receptor β in Alzheimer's disease: from mechanisms to therapeutics. Ageing ResRev. 2015;24(B):178-90. doi: 10.1016/j.arr.2015.08.001, PMID 26307455.
63. Khosla S, MeltonIII LJ, Atkinson EJ, O'fallon WM. Relationship of serum sex steroid levels to longitudinal changes in bone density in young versus elderly men. JClinEndocrinolMetab. 2001;86(8):3555-61. doi: 10.1210/jcem.86.8.7736, PMID 11502778.
64. Lan YL, Zhao J, Li S. Estrogen receptors' neuroprotective effect against glutamate-induced neurotoxicity. NeurolSci. 2014;35(11):1657-62. doi: 10.1007/s10072-014-1937-8, PMID 25228013.
65. Zárate S, Stevnsner T, Gredilla R. Role of estrogen and other sex hormones in brain aging. Neuroprotection and DNA repair. FrontAgingNeurosci. 2017;9:430. doi: 10.3389/fnagi.2017.00430, PMID 29311911.
66. Pompili A, Arnone B, Gasbarri A. Estrogens and memory in physiological and neuropathologicalconditions. Psychoneuroendocrinology.2012;37(9):1379-96. doi: 10.1016/j.psyneuen.2012.01.007, PMID 22309827.
67. C Correia S, X Santos R, Cardoso S, Carvalho C, S Santos M, R Oliveira C, I Moreira P. Effects of estrogen in the brain: is it a neuroprotective agent in Alzheimer's disease? Current aging science. 2010; 3(2):113-26.
68. Xu H, Gouras GK, Greenfield JP, Vincent B, Naslund J, Mazzarelli L, Fried G, Jovanovic JN, Seeger M, Relkin NR, Liao F, Checler F, Buxbaum JD, Chait BT, Thinakaran G, Sisodia SS, Wang R, Greengard P, Gandy S. Estrogen reduces neuronal generation of Alzheimer β-amyloid peptides. NatMed. 1998;4(4):447-51. doi: 10.1038/nm0498-447, PMID 9546791.
69. Tota S, Nath C, Najmi AK, Shukla R, Hanif K. Inhibition of central angiotensin converting enzyme ameliorates scopolamine induced memory impairment in mice: role of cholinergic neurotransmission, cerebral blood flow and brain energy metabolism. Behav Brain Res.2012;232(1):66-76. doi: 10.1016/j.bbr.2012.03.015, PMID 22460064.
70. Hancianu M, Cioanca O, Mihasan M, Hritcu L. Neuroprotective effects of inhaled lavender oil on scopolamine-induced dementia via anti-oxidative activities in rats. Phytomedicine. 2013;20(5):446-52. doi: 10.1016/j.phymed.2012.12.005, PMID 23351960.
71. Liem-Moolenaar M, de Boer P, TimmersM, Schoemaker RC, van Hasselt JG, Schmidt S, van Gerven JM. Pharmacokinetic-pharmacodynamic relationships of central nervous system effects of scopolamine in healthy subjects. BrJClinPharmacol. 2011;71(6):886-98. doi: 10.1111/j.1365-2125.2011.03936.x, PMID 21306419.
72. Kwon SH, Ma SX, Joo HJ, Lee SY, Jang CG. Inhibitory Effects of Eucommia ulmoidesOliv. Bark on scopolamine-induced Learning and memory Deficits in Mice. BiomolTher (Seoul). 2013;21(6):462-9. doi: 10.4062/biomoltther.2013.074, PMID 24404337.
73. Souza AC, Bruning CA, Acker CI, Neto JS, Nogueira CW. 2-Phenylethynyl-butyltellurium enhances learning and memory impaired by scopolamine in mice. BehavPharmacol.2013;24(4):249-54. doi: 10.1097/FBP.0b013e32836353a5, PMID 23751517.
74. Kim SJ, Lee JH, Chung HS, Song JH, Ha J, Bae H. Neuroprotective Effects of AMP-Activated Protein Kinase on Scopolamine Induced Memory Impairment. Korean JPhysiolPharmacol. 2013;17(4):331-8. doi: 10.4196/kjpp.2013.17.4.331, PMID 23946693.
75. Azami NS, Jahanshahi M, Babapour V. The role of CA1 α-adrenoceptor on scopolamine induced memory impairment in male rats. PhysiolPharmacol. 2010;14(1):66-77.
76. Bhattacharya A, Muruganandam AV, Kumar V, Bhattacharya SK. Effect of poly herbal formulation, EuMil, on neurochemical perturbations induced by chronic stress. Indian JExpBiol. 2002;40(10):1161-3. PMID 12693697.
77. Parihar MS, Hemnani T. Experimental excitotoxicity provokes oxidative damage in mice brain and attenuation by extract of *Asparagus racemosus*. JNeural Transm (Vienna). 2004;111(1):1-12. doi: 10.1007/s00702-003-0069-8, PMID 14714211.
78. Nandgopoal M, Muralidharan P, Thirumurugan G. Cerebroprotective effect of root extract of *Asparagus racemosus*Wild in global cerebral ischemia in rats. JPharmacolToxicol. 2011;6(1):49-61.
79. Laddawan L, Hathairal K, Patcharada A, Onrawee K. Neuroprotective effects of the *Asparagus racemosus* root extract on ovariectomized rats. Journal of Physiological and Biomedical Scienc. 2013;26(1):18-22.
80. Hayes PY, Jahidin AH, Lehmann R, Penman K, Kitching W, De Voss JJ. Steroidal saponins from the roots of *Asparagus racemosus*. Phytochemistry. 2008;69(3):796-804. doi: 10.1016/j.phytochem.2007.09.001, PMID 17936315.
81. Ojha R, Sahu AN, Muruganandam AV, Singh GK and Krishnamurthy S. Asparagus racemosus enhances memory and protect against amnesia in rodent models. Brain and Cognition.2010; 74(1): 1-9.

Cite this article: Jagdish P, Maheep B. Neuroprotective Efficacy of *Asparagus racemosus* Root Extract on Hippocampal Neurons in Scopolamine Mouse Model of Alzheimer's Disease. Asian J Biol Life Sci. 2021;10(2):391-9.