EVALUATION OF THE POSSIBLE ANTIOXIDANT EFFECTS OF PEGANUM HARMALA AND GINKGO BILOBA IN AMELIORATING ALZHEIMER'S DISEASE IN RAT MODEL

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ABSTRACT: Alzheimer's disease (AD) is a progressive and irreversible neurodegenerative disorder leading to dementia in the elderly inhabitants. Increasing evidence advises that oxidative stress that is normally associated with aging is an obvious and early feature of AD and plays a role in its pathogenesis. The present study was designed to evaluate the possible neuroprotective activity of Ginkgo biloba (Gb) and / or Peganum harmala (Ph) extract against AD in a rat model. AD was induced by chronic administration intraperitoneal injection of scopolamine (0.7 mg kg⁻¹, i.p.) to rats for a period of 7 days. The Gb (120 mg/kg b.wt.) and Ph (187.5 mg/kg b.wt.) were administrated to AD rat group orally daily for a period of 30 days. The results revealed that the levels of thiobarbituric acid reactive substances and Xanthine oxidase were significantly increased, even though the activities of superoxide dismutase and catalase, as well as the reduced glutathione, were significantly decreased in the brain homogenate of Alzheimer group. Additionally, brain acetylcholinesterase as well as alkaline phosphatase, acid phosphatase, alanine aminotransferase, aspartate aminotransferase and lactate dehydrogenase activities were significantly increased. On the other hand, the administration of Gb, Ph and co-administration of those test herbs acquired potential therapeutic effects on improving the neurodegenerative disorder in rats through suppressing lipid peroxidation, augmenting endogenous antioxidant enzymes, and reducing acetylcholinesterase activity in the brain. It might be concluded that a mixture extract, by its antioxidant constituents, could modulate scopolamine-induced oxidative stress and enzyme activities in the brain.

INTRODUCTION: Alzheimer’s disease is a pathological brain disease and the most frequent cause of dementia 1,2. Patients with AD suffer a gradual deterioration of memory and other cognitive functions, which eventually causes a complete incapacity and fatal within 3 to 9 years after diagnosis 3. Aging is the significant hazard factor of AD in the general population, it influences considerably on the everyday activities of older adults, being one of the key factors behind disability in older age. AD affects one person in eight over sixty-five and almost half over eighty five years of age, it is the 5th leading cause of death of the elder people 4. There was clearly a predicted 46.8 million worldwide people developed dementia, and the incidence is expected to rise in the coming years, with 74.7 million cases approximated to occur in 2030 and 131.5 million in 2050 5. The etiology of AD is multifactorial, including genetic, environmental, and lifestyle.
Several other factors such as absence of acetylcholine, the development of atherosclerotic lesions and inflammation of the brain - also play an important role in the progression of AD. Albeit AD is most likely connected with various etiologies and pathophysiologic mechanisms, oxidative stress looks as a noteworthy area of the pathophysiologic process.

Oxidative stress (OS), a process increased in the brain with aging, is induced by an imbalance in the redox state, including the generation of abnormal reactive oxygen species (ROS) or the dysfunction of the antioxidant system. These ROS assume a part in neurodegenerative diseases.

There has been an expanding enthusiasm for as far back as decades about intercessions that may enhance cognitive performance in older age or, in any event, defer the beginning of dementia. Because of the nonattendance of a cure against dementia and AD, the public health concern has concentrated more lately on banning of cognitive decline. Lately, the role of food and nutrition in preventing or delaying chronic impairment in the elderly population has brought a great attention, because of their ability to influence biochemical and biological processes, bioactive nutrients are considered flexible factors capable of conserving a healthy brain status.

*Ginkgo biloba* (Gb) has long been considered to have therapeutic properties, and its extracts are presently the most investigated and an adopted herbal treatment for cognitive disorders and AD, and it is summarized as an antidementia medication in the Anatomical Treatment Chemical Classification system. In addition, *in vitro* and *in vivo* studies have reported that Gb plays a key role in the maturation and development of neuro-fibrillary tangles. From medical trials with Gb, it is often estimated that Gb delays progression of symptoms in Alzheimer’s dementia by about 6 - 9 months. Extract of Gb contains flavonols quercetin, kaempferol as well as terpenes that’s giving this extract its unique polyvalent pharmacological action for use as a neuroprotective function. It is well known due to the anti-inflammatory, anti-apoptotic and antioxidant properties which contribute its ability to scavenge free radicals.

Earlier investigation reported the essential prophylactic role of Gb in the prevention of several diseases associated with oxidative tissue damage. *Peganum harmala* (Ph) belongs to the family of Zygophyllaceae. An assortment of pharmacological and biological activities of Ph inclusive of antibacterial, anti fungal and monoamine oxidase (MAO) inhibition has been mentioned. This herb is wealthy in alkaloids; β-carbolines including harmalol, harmaline, norharmane, harmol, harmine and harmane. The harmine alkaloids exhibit in *Peganum harmala* have been accounted for to sufficiently supportive to utilize in the management of neurodegenerative disorders of the type Alzheimer’s diseases through its acetylcholinesterase (AChE) inhibitory activity. The aqueous extract of *Peganum harmala* should save your signs and symptoms and reduced oxidative stress markers in rats with Parkinson’s disease caused by 6-hydroxydopamine.

The current study was planned to ascertain the possible neuroprotective activity of *Peganum harmala* and *Ginkgo biloba* extract administered individually or in combination, in safeguarding the brain against the Alzheimer’s disease in a rat model.

**MATERIALS AND METHODS:**

**Experimental Animals:** Male Wistar rats obtained from the Animal house of King Fahad clinical research Center, Jeddah, Kingdom of Saudi Arabia, weighing 220 - 250 g, have been used. The animals preserved under preferred situations of temperature and humidity and a 12-h mild dark cycle with access to food and water for one-week *ad libitum* prior to experimentation. Animals were cared for in accordance with the principles of the “Guide to the Care and Use of Experimental Animals” (Committee on the Care and Use of Laboratory Animals 1985) and by King Fahd Research Center, King Abdul-Aziz University.

**Induction of Experimental Alzheimer’s Disease:** Scopolamine hydrochloride was purchased from Sigma -Aldrich, St, Louis, MO, USA. Scopolamine was solved with a 0.9% saline solution and being injected in a volume of (0.7 mg / kg, i.p.) of body weight to rats for a period of 7 days.
Plant Material:
**Ginkgo biloba**: Ginkgo biloba capsules were used in this study, each capsule containing (260) mg of ginkgo leaves manufactured by EIMC United Pharmaceuticals in Egypt with M.O.H Reg. no. 589/2012. Each capsule was solved in 3 ml of distilled water. The drug was given daily (120 mg/kg body weight) by oral gavage syringe for 30 days.

**Peganum harmala**: Dry plant seeds of Peganum harmala were obtained from the herbal market in Cairo, Egypt. The crude extract was obtained according to the method Al-Izzy

Preparation of Extract: Seeds of Ph were purified washed and dried under fresh air, then ground in an electrical grinder to get fine powder of the seeds. Then, 100 gm of the ground seeds was infused in 500 ml of distilled water for 24 hours at room temperature. Agitation of the infusion by using a magnetic stirrer had been done alternatively. Then the infusion was filtered by filter paper (Wattman no.1) and the deposit was discarded. The extract was administrated to rats at doses (187.5 mg/kg b.wt.) daily by oral gavage syringe for 30 days.

The Experimental Design: 50 male Wistar rats were randomly assigned to five groups of ten animals each as follows:

**Group 1**: (C) Normal, healthy rats served as an untreated negative control group.

**Group 2**: (AD) rats received Scopolamine hydrochloride (0.7 mg kg⁻¹, i.p.) for 7 days to induce AD and served as an untreated positive control group.

**Group 3**: (AD + Gb) rats received Scopolamine (0.7 mg kg⁻¹, i.p.) and treated orally with Ginkgo biloba (120 mg/kg b.wt.) daily for 30 days.

**Group 4**: (AD + Ph) rats received Scopolamine (0.7 mg kg⁻¹, i.p.) and treated orally with Peganum harmala (187.5 mg/kg b.wt.) daily for 30 days.

**Group 5**: (AD + Ph + Gb) rats received Scopolamine (0.7 mg kg⁻¹, i.p.) and treated orally with mixture of Peganum harmala (187.5 mg/kg b.wt.) and Ginkgo biloba (120 mg/kg b.wt.) daily for 30 days.

Methods:
**Measurement of Body Weight (Biological Evolution)**: The rats in all groups were monitoring the change in body weight by measuring their weight before the start of treatment and every week during the experimental period.

**Brain Tissue Sampling and Preparation**: At the end of the experimental duration, the rats were sacrificed by using decapitation after an overnight fast of 12 hours. The whole brain of each rat was rapidly removed, washed with ice-cold saline, blotted dry and weighed. Then, the brain become perfused with a PBS (phosphate buffered saline) solution, pH 7.4. Containing 0.16 mg/ml heparin to remove any red blood cells and clots. Then homogenize the tissue in 5 - 10 ml cold buffer (i.e., 50 mM potassium phosphate, pH 7.4. 1 mM EDTA and 1 mL/L Triton X-100) per gram tissue. The homogenate was centrifuged at 4,000 rpm for 15 minutes at 4 °C and the resultant supernatant was used for different determinations.

**Biochemical Analysis**: Brain acetyl cholinesterase (AChE), lactate dehydrogenase (LDH), alkaline phosphatase (ALP), acid phosphatase (AP), Aspartate Aminotransferase (AST) and Alanine Aminotransferase (ALT) activities as well as, lipid peroxidation assay (TBARS), Xanthine oxidase (XO), reduced glutathione estimation (GSH), superoxide dismutase (SOD) and catalase (CAT) were estimated according to the protocol mentioned in the assay kits supplied by the biodiagnostic Chemical Company (Egypt).

**Statistical Evaluation**: All values had been expressed as mean ± standard deviation (X ±SD) obtained from the experiments (n = 10). Statistical analysis became performed with one-way analysis of variance (ANOVA) test and independent pattern t-test using the Mega Stat Excel (version 10.3, Butler University).

**RESULTS**: As shown in Table 1, the body weights (BWTs) and brain weight of the rats in different treatment groups were matched to the mean value within the normal group. It was observed a significant decrease in the final body weight of the Alzheimer group compared with normal group. However, no significant decreased had been found in brain weights as compared with
a normal group. Treating of AD group with either Gb and /or Ph exhibits significant improvement of the body weight in comparison with untreated AD group.

**TABLE 1: INITIAL, FINAL BODY WEIGHT (g) AND BRAIN WEIGHT, IN CONTROL AND AD RATS TREATED WITH GINKGO BILOBA, PEGANUM HARMALMA OR THEIR MIXTURE AFTER 4 WEEKS OF TREATMENT**

<table>
<thead>
<tr>
<th>Experimental groups</th>
<th>Initial body weight (g)</th>
<th>Final body weight (g)</th>
<th>Brain weight (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>235.00 ± 7.517</td>
<td>277.60 ± 26.264</td>
<td>1.800 ± 0.065</td>
</tr>
<tr>
<td>AD</td>
<td>237.40 ± 6.148</td>
<td>249.20 ± 22.643**</td>
<td>1.620 ± 0.369</td>
</tr>
<tr>
<td>AD + Gb</td>
<td>239.00 ± 5.000</td>
<td>267.50 ± 17.111</td>
<td>1.763 ± 0.092</td>
</tr>
<tr>
<td>AD + Ph</td>
<td>239.60 ± 3.507</td>
<td>262.20 ± 24.576</td>
<td>1.843 ± 0.092</td>
</tr>
<tr>
<td>AD + Gb-Ph</td>
<td>240.60 ± 4.159</td>
<td>274.40 ± 23.776</td>
<td>1.836 ± 0.098</td>
</tr>
</tbody>
</table>

Each value represents the mean of 10 rats ± SD. There is significantly different from control values at 0.01**.

Regarding antioxidants, the data indicated that, AD induced a significant reduction in brain SOD and CAT activities as well as GSH content in comparison to a control group Table 2. However, treated AD group with the aqueous extract of Gb or Ph compared to the untreated AD group. The level of these enzymes was significantly suppressed by the administration of rats with Gb or Ph compared to the AD value at P < 0.05*, 0.01**, 0.001***. There is significantly different from the AD value at P < 0.05*, 0.01**, 0.001***

**TABLE 2: THE EFFECT OF THE AQUEOUS EXTRACT OF GB OR / AND PH ON BRAIN SOD, CAT ENZYME ACTIVITY AND GSH LEVELS IN SCopolamine Induced AD RATS**

<table>
<thead>
<tr>
<th>Groups</th>
<th>GSH (M mol/g)</th>
<th>CAT (µ/g)</th>
<th>SOD (µ/gm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>0.553 ± 0.088</td>
<td>0.404 ± 0.046</td>
<td>330.688 ± 80.646</td>
</tr>
<tr>
<td>AD</td>
<td>0.380 ± 0.080***</td>
<td>0.168 ± 0.046 ***</td>
<td>148.241 ± 24.370***</td>
</tr>
<tr>
<td>AD + Ph</td>
<td>0.483 ± 0.071*</td>
<td>0.300 ± 0.050*</td>
<td>297.780 ± 72.380***</td>
</tr>
<tr>
<td>AD + Gb</td>
<td>0.446 ± 0.034***</td>
<td>0.320 ± 0.151###</td>
<td>284.891 ± 83.966***</td>
</tr>
<tr>
<td>AD + Gb-Ph</td>
<td>0.505 ± 0.146###</td>
<td>0.371 ± 0.093###</td>
<td>301.185 ± 84.354###</td>
</tr>
</tbody>
</table>

Data were expressed as X ± SD. There is significantly different from control values at P < 0.05*, 0.01**, 0.001***. There is significantly different from the AD value at P < 0.05*, 0.01**, 0.001***

The data presented in Table 3, showed that the XO and MDA levels were significantly increased in (P-value = 0.000) in AD group when compared to the normal group. Treatment of AD group with Gb or Ph ameliorated the levels of XO and MDA by reducing them significantly relative to the untreated AD group.

**TABLE 3: THE EFFECT OF THE AQUEOUS EXTRACT OF GB OR / AND PH ON THE OXIDATIVE STRESS IN THE BRAIN OF THE EXPERIMENTAL GROUPS**

<table>
<thead>
<tr>
<th>Groups</th>
<th>XO (µ/ml)</th>
<th>MDA (N mol/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>25.172 ± 4.348</td>
<td>15.003 ± 1.728</td>
</tr>
<tr>
<td>AD</td>
<td>37.282 ± 7.457***</td>
<td>24.167 ± 5.021***</td>
</tr>
<tr>
<td>AD+ Ph</td>
<td>29.672 ± 2.052^##</td>
<td>18.603 ± 4.773##</td>
</tr>
<tr>
<td>AD + Gb</td>
<td>28.433 ± 4.002^###</td>
<td>18.360 ± 1.075##</td>
</tr>
<tr>
<td>AD+ Gb-Ph</td>
<td>23.785 ± 2.241###</td>
<td>17.755 ± 4.544###</td>
</tr>
</tbody>
</table>

Data were expressed as X ± SD. There is significantly different from control values at P < 0.05*, 0.01**, 0.001***. There is significantly different from the AD value at P < 0.05*, 0.01**, 0.001***

It is clear from the results that injection of rats with scopolamine caused a significant elevation (P-value = 0.000, 0.000, 0.002) in the activity of AChE, LDH and AP respectively compared to that of the control group. The level of these enzymes was significantly suppressed by the administration of rats with Gb or Ph compared to the untreated AD group Table 4. As regards to AST and ALT enzyme, the data demonstrated in Table 5 showed there were highly significant increases (P-value = 0.000) in their activities in AD group when compared to the normal group. In other hand, the activity of ALP enzyme in AD group was slightly significantly elevated relative to the normal group. The results of the current study showed a significant attenuation of the Gb or / and Ph, effects on AST and ALT activity in the AD groups by decreasing it significantly when compared to the untreated AD group. Whilst, AD groups treated with either Gb nor Ph showed non-significant change in ALP activity when compared to the untreated AD group.
TABLE 4: THE EFFECT OF AQUEOUS EXTRACT OF GB OR / AND PH ON BRAIN ACETYLCHOLINESTERASE, LACTATE DEHYDRGENASE AND ACID PHOSPHATASE ACTIVITIES IN SCOPOLAMINE-INDUCED AD RATS

<table>
<thead>
<tr>
<th>Groups</th>
<th>AChE µ/mg</th>
<th>LDH µ/L</th>
<th>AP µ/L</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>411.904 ± 48.83</td>
<td>13.395 ± 2.41</td>
<td>6.163 ± 1.28</td>
</tr>
<tr>
<td>AD</td>
<td>539.901 ± 45.411</td>
<td>40.532 ± 9.524</td>
<td>10.416 ± 2.496</td>
</tr>
<tr>
<td>AD + Ph</td>
<td>473.990 ± 52.407</td>
<td>24.922 ± 7.050</td>
<td>7.298 ± 2.180</td>
</tr>
<tr>
<td>AD + Gb</td>
<td>463.056 ± 46.840</td>
<td>23.058 ± 5.904</td>
<td>7.210 ± 3.524</td>
</tr>
<tr>
<td>AD + Ph-Gb</td>
<td>447.331 ± 60.483</td>
<td>19.910 ± 4.986</td>
<td>7.139 ± 1.891</td>
</tr>
</tbody>
</table>

Data were expressed as X ± SD. There is significantly different from control values at P <0.05*, 0.01** 0.001***. There is significantly different from the AD value at P <0.05*, 0.01** 0.001***

TABLE 5: THE EFFECT OF THE AQUEOUS EXTRACT OF GB OR / AND PH ON THE BRAIN ENZYME ACTIVITIES IN SCOPOLAMINE-INDUCED AD RATS

<table>
<thead>
<tr>
<th>Groups</th>
<th>AST (µ/L)</th>
<th>ALT (µ/L)</th>
<th>ALP (µ/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AD</td>
<td>39.053 ± 7.454</td>
<td>25.815 ± 4.960</td>
<td>560.459 ± 254.092*</td>
</tr>
<tr>
<td>AD + Ph</td>
<td>22.322 ± 2.232</td>
<td>16.947 ± 2.076</td>
<td>436.262 ± 67.228</td>
</tr>
<tr>
<td>AD + Gb</td>
<td>25.124 ± 3.265</td>
<td>17.672 ± 2.260</td>
<td>408.561 ± 150.492</td>
</tr>
<tr>
<td>AD + Ph-Gb</td>
<td>24.124 ± 1.863</td>
<td>14.778 ± 2.700</td>
<td>396.614 ± 160.867</td>
</tr>
</tbody>
</table>

Data were expressed as X ± SD. There is significantly different from control values at P <0.05*, 0.01** 0.001***. There is significantly different from the AD value at P <0.05*, 0.01** 0.001***

DISCUSSION: There is growing evidence that AD became a major medical, economic, and social problem that is deteriorating as the increasing number of elderly people. The increasing in brain oxidative stress during brain aging could be reversed by antioxidants. Currently, available drug therapies for Alzheimer’s disease and other diseases that because dementia consists primarily of acetylcholinesterase inhibitors and some neuroprotective agents. These drugs cause side effects, thus an alternative bioactive compound from plants with little or no side effects should be used to replace these drugs. Therefore, the present research was undertaken to check out the effects of Gb and Ph, (both individual and combinational) in the scopolamine-treated rat model of Alzheimer.

The present results demonstrate that there was a decrease in SOD, CAT activities and GSH level with the concomitant increase in MDA and XO levels in the brain of AD group as compared to the normal group. This finding is in accordance with the results of Various specialized clinical research has reported strong evidence that oxidative stress is included in the pathogenesis of Alzheimer’s disease. The susceptibility of brain to oxidative stress other than many tissues is caused by the neuronal membranes that comprise a very excessive percentage of long chain polyunsaturated fatty acids. Consequently, lipids consider as one of the targets of oxidative change by way of free radicals in neurodegenerative disorders. The elevated oxidative stress in brains can be by inducing the activation of a cascade of redox-sensitive cell signal pathways. This likely displays the increase in lipid peroxidation due to either an increase in the production of free oxidative radicals or a decrease in the antioxidant defense mechanisms such as α-tocopherol as a result directing the role of free radicals in the progression of Alzheimer's disease.

The current results demonstrated that XO activity was significantly increased in AD group when compared with the normal group. These results are in coincide with. They increased in brain XO activity may be contributing to the increase in intracellular calcium, which activates proteases that catalyze conversion of xanthine dehydrogenase to xanthine oxidase, which in turn catalyzes purine bases to form O2-. Therefore, diminished energy metabolism can increase intraneuronal calcium, which leads to excitotoxicity, and these converging mechanisms are able to be producing ROS. Each of these mechanisms are thought to occur in neurodegenerative diseases, mainly AD. The depletion of brain GSH in Alzheimer rat model has been reported previously by.

GSH is the most plenteous endogenous antioxidant in the brain, which found mainly in the reduced form within the cells. Therefore, it has been shown to react with free radicals and prohibit generation of hydroxyl free radicals. The decreased level of GSH in scopolamine- treated animals indicates...
that there is an increased generation of free radicals and reduced activity of glutathione system in fighting oxidative stress 45.

Catalase performs a major role in detoxifying superoxide anions, which furthermore damages the cell walls and macromolecules 48. The decreased in the catalase activity in scopolamine - induced AD group is accompaniments with other studies 31,49,50. Marisco 51 and Ishola 52 deduced that Scopolamine has been found to increase oxidative stress and impaired the anti-oxidative defense system.

The decreased activity of SOD may be due to the toxic effect of Scopolamine by promoting oxidative stress in the brain and this may affect brain functioning and growth. Moreover, this decrease may be as a result of imbalances between oxidants and antioxidant level in support of the oxidants 49, 53. This result is steady with the study of Otitoju 55.

The data obtained from the present study mentioned that the administration of a normal water extract of Gb and / or Ph to scopolamine stimulate AD rats caused a significant decrease in the level of TBARS, XO and elevated the SOD and CAT enzyme activities and GSH contents when compared with the untreated AD group.

The antioxidative effects of Ph were studied by Rezaei 29, who concluded that the aqueous extract of Peganum harmala could prevent signs and decreased oxidative stress markers in rats with Parkinson’s disease brought on by 6-hydroxydopamine. Hamden 55 decided that, Ph contains effective constitutes which can also have the ability of scavenging free radicals and modulate the expression of genes encoding antioxidant enzymes inclusive of estrogens, growth factors and Vitamin E supplement. Moreover, the alkaloid extract of seeds of Ph has a prospect enough at the control of neurodegenerative disorders of the type Alzheimer’s diseases 27. Farzin and Mansouri 56 indicated that β-carbolines (harmane, norharmane and harmine) induce an antidepressant-like impact, while, harmaline and harmane are capable of decrease voltage-gated calcium channel currents at concentrations which can be can be to be sufficient for neuroprotective effects in vivo.

Concerning the possible ameliorative role of Ginkgo biloba, the present results revealed that the free radical scavenging and antioxidant activity of Gb is confirmed via restoration of XO and MDA levels along with a significant increase of CAT and SOD activities and GSH level in the brain as compared with the values recorded in normal rats. These results agree with other previous findings 57, 58. This probably due to biologically active ingredients which are rich in Gb 59, 60. Further, Ginkgo leaves extract is identified to prevent the creation of Aβ from a β-amyloid precursor protein (APP), a critical method within the pathogenesis of Alzheimer's disease 61. Alternatively, the Gb inhibits ROS accumulation induced by Aβ (especially flavonol quercitin) and also reduces neuron apoptosis, in which apoptosis is considered to be one of the major causes for neurodegenerative diseases. 62 suggested that the neuroprotective effects of Gb can be due to their antioxidant properties, antiapoptotic effects, and improvement of energy metabolism.

The present study confirmed that scopolamine caused AD produced a significant increase in AChE activity in contrast with the control group. This finding is in accordance to the results of 63, 65. The preserving of the acetylcholine level within the brain is important in the proper healthcare of AD patients. One particular method in that is blocking the activity of AChE, the enzyme-degrading acetylcholine 60.

On the other side, AChE activity in AD rats treated with Ph and Gb, (both individual and combinational) was inhibited. It absolutely was documented that Gb promote the ability of learning and memory by increasing Ach level and inhibiting AChE activity 67, 68. Regard the Ph, previous reviews have noted that Ph indicates a potential therapeutic effect on AD, because of the cholinesterase inhibitory activities of harmine, harmaline, harmalol, harmol, and vasicine presented in plant 69, 71. Further, Singh 72 explained that the deoxypeganie from harmala seeds has activity similarly to reversibly behaving cholinesterase inhibitors. It suppresses acetylcholinesterase and monoamine oxidase, thereby protecting against the degradation of acetylcholine and dopamine, which is therefore beneficial in treating Alzheimer’s dementia.
One of the major findings of the present study was the elevation of the activity of brain LDH in Alzheimer's group. These results are relative to the findings of 73 - 75. LDH serves as an essential metabolic enzyme in brain cells and it is released into the bloodstream from injuries brain cells. Thus, LDH level in serum or brain cell is a reliable index to assess cerebral ischemic injury 76. Elevated extracellular LDH is a biomarker of oxidative stress and disrupted cell sincerity throughout the lipid peroxidation and oxidative stress.

Tao 74 reported that increased level production of LDH cause Aβ accumulation is probably involved in the pathophysiology of AD. Administration of Gb and Ph to AD groups caused a pronounced reduction in the elevated activity of LDH as compared with Alzheimer’s group. Much of those decrease could be due to the antioxidant characteristic in their constituents as flavonoids and terpenoids in Ginko and harming alkaloids from the seeds of the, which protect the cellular membrane probity from scopolamine-induced oxidative damage and restore the antioxidant system, therefore, enhance brain function.

As shown in a present study, scopolamine increased the brain AP activity in the AD group as compared with control rat group. These findings are in agreement with some studies reported by 77, 78. Change in the activity of AP causes Lysosomal storage disorders 79 and neurodegenerative diseases 80. Moreover, dysfunction lysosome system results in the formation of Tau insoluble aggregates in lysosomes of AD 81. A significant reduction in brain AP activity was recorded in AD group treated with Gb and /or Ph in the present study as compared to the untreated AD group. The effect of Gb in reducing AP agrees with Tan 82. In addition, Mohamed and El-Moneim 83, observed an improvement in AP activity in rats administrated of AlCl3 after treatment with Gb.

In the present study, a significant increase in brain AST, ALT, and ALP activities were observed in the AD untreated group as compared to control rats. These results are in accordance with the findings of Sumathi 84 who noticed an increased in the activities of ALP, ACP, ALT and AST in all the brain parts of aluminium induced neurotoxicity group when compared with control rats. AST and ALT are important enzymes of brain; their activities are related to the maintenance of amino acid homeostasis and might be an indicator of mitochondrial injury 85. Brain is enriched in amino compounds, it is possible that the promote transamination activity within the brain tissue served to neutralize the biochemical and biological responses that happen due to the amino compounds in response to the scopolamine effects 86. Treated AD+ group with the water extract of Gb and /or Phameliorate the enzyme activities which were brought back close to normal compared to the untreated AD+ group.

This result indicates that Gb extract may play important roles in the improvement of the brain of the Alzheimer's disease through augmentation of bloodstream flow and inhibition of platelet activating factor, it may also protect the cell membrane against injury induced by free radicals and has protective effects against brain ischemia / reperfusion injury 87. Moreover, the protective impact of Ph extract may be related to the richness of the plant in substances of phenolic characteristics which may decrease the free - radical lipid peroxidation level leading to the stabilizing of membrane structures 55.

CONCLUSION: In conclusion, the present study revealed that Peganum harmala and Ginkgo biloba extracts possibly have a protective role against AD by inhibiting oxidative stress, increase the antioxidant enzymes and improve the alteration of enzyme activities in the brain. Therefore, the receipt of these herbs may encourage older people to improve general health condition.

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CONFLICT OF INTEREST: The authors declare no conflict of interest associated with this work.

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