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ORIGINAL ARTICLE

# Effect of *Nigella sativa* and wheat germ oils on scopolamine-induced memory impairment in rats

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**KEYWORDS**

*Nigella sativa* oil;
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Memory impairment;
Rats

**Abstract** *Aim:* To investigate the possible memory enhancing effects of *Nigella sativa* oil (NSO) and wheat germ oil (WGO) on scopolamine-induced amnesic rats.
*Methods:* Male Wistar rats received either saline or scopolamine (16 mg/kg, i.p.). The other three groups were pretreated with NSO (1 ml/kg, p.o.), WGO (170 mg/kg, p.o.) or donepezil used as a reference drug (10 mg/kg, p.o.) for 14 days before scopolamine injection. Cognitive and biochemical measurements were then assessed.
*Principal results:* NSO and WGO treated rats significantly reversed scopolamine-induced deficit of spatial and non-spatial working memory impairment in the T maze alternation task and object recognition test, respectively. Administration of NSO prior to scopolamine showed a significant decrease in malondialdehyde (MDA) and increase in Glutathione (GSH) brain contents to be similar to that observed in donepezil group. It did not alter cholinesterase activity and showed a significant decrease in brain tumor necrosis factor-alpha (TNF-α) content to be similar to donepezil-treated rats. Scopolamine-demented rats pretreated with WGO did not change MDA brain content significantly as compared to scopolamine and donepezil groups. WGO-treated rats showed a significant increase in GSH to a level similar to that observed in the donepezil group, it showed a significant decrease in cholinesterase activity as compared to scopolamine group and significantly elevated brain TNF-α content when compared to donepezil group.

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*Conclusions:* Memory enhancing effect of NSO in the present study might be due to its antioxidant and anti-inflammatory activities, while that of WGO might be via its antioxidant and anticholinesterase activities.



## 1. Introduction

Dementia is characterized by a loss of or decline in memory and other cognitive abilities. Alzheimer's disease (AD) is one of the most common subtypes of dementia.[1] It is characterized by deposition of amyloid plaques, neurofibrillary tangles, cerebral oxidative stress, inflammation and impaired neuronal function.[2]

The operant mechanism of AD in the elderly population is related to cholinergic system dysfunctions such as the loss of cholinergic neurons in the basal forebrain and hippocampus.[3] Acetylcholine functions as the principal neurotransmitter and its decreased release also results in learning deficits.[4] Both acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) are best known to hydrolyze acetylcholine rapidly.[5] In patients with AD the level of AChE activity declines and the activity of BChE increase in certain brain regions. Cholinesterase inhibition is the most used treatment for the symptoms of AD and AChE as well as BChE are therapeutic targets for improving the cholinergic deficit.[6]

Scopolamine, a muscarinic receptor antagonist, have profound amnesic effects in a variety of learning paradigms and is considered a useful experimental approach to investigate the mechanisms involved in cognitive impairment observed in AD.[7] Scopolamine also mediates cholinergic deficit through oxidative stress[8] and neuroinflammation.[9]

Acetylcholinesterase inhibitors (AChEIs) e.g. donepezil, rivastigmine and galantamine, are the most effective pharmacotherapeutic agents for AD. However, these agents also cause undesirable side effects that limited their use.[10] Donepezil also suppressed oxidative stress in the animal model of dementia.[11] Furthermore, it exerts its therapeutic effect in AD not only by inhibition of AChE but also by its anti-inflammatory effect.[12,13]

NSO has been extensively reported to exhibit protective effect against many diseases depending on its high antioxidant[14] as well as anti-inflammatory activities.[15] Moreover, reactive oxygen species are implicated in the mechanism of several neurodegenerative diseases such as AD[16] therefore NSO could be used as a possible remedy for cognitive disorders.

WGO has the highest tocopherol content of all vegetable oils, and particularly the highest content of α-tocopherol (vitamin E), which represents around 60% of the total content.[17] Vitamin E is a potent antioxidant agent that has been described to protect the brain from oxidative stress by directly scavenging toxic radicals[18] and ameliorate the brain impairment process underlying aging.[19] Moreover brain AChE was markedly reduced by supplementation with vitamin E.[20]

The present study was constructed in order to assess the influence of NSO and WGO on behavioral alterations in demented rats. Donepezil was used as a reference drug. Moreover, their effects on brain oxidative stress indices, cholinergic deficit and inflammation were also evaluated.

## 2. Materials and methods

### 2.1. Animals

Male albino Wistar rats weighing 120–150 g were used throughout the experiment. They were obtained from the animal house colony of the National Research Centre (Dokki, Cairo, Egypt) and were housed for at least one week in the laboratory room prior to testing under standard housing conditions. Animals were fed standard laboratory pellets with water *ad libitum*. All animal procedures were performed in accordance with the Ethics Committee of the National Research Centre, Egypt. Registration number (09/189).

### 2.2. Drugs and chemicals

Scopolamine hydrobromide was purchased from Sigma–Aldrich (MO, USA) and was dissolved in saline (0.9% NaCl). Donepezil hydrochloride was purchased from Pfizer (Giza, Egypt) and was freshly prepared in 1% tween 80 in water. NSO and WGO were obtained as a gift from Mepaco (Sharkia, Egypt). WGO was used as 1% v/v oil in water emulsion.

### 2.3. Treatments

Rats were randomly allocated into five groups (10 rats/group in the T maze alternation task and 8 rats/group in the object recognition test) as follows: group I received saline and served as normal while group II received scopolamine (16 mg/kg, i.p.)[21] and served as control. Both groups received saline for 14 days. Groups III–V received donepezil (10 mg/kg, p.o.),[22] NSO (1 mg/kg, p.o.)[23] (equivalent to 0.92 mg/kg) or WGO (170 mg/kg, p.o.),[24] respectively for 14 days. Scopolamine was administered as a single dose 30 min after the last administration in groups II–V.

### 2.4. Cognitive measurements

#### 2.4.1. T maze alternation task

The spatial alternation maze was designed as described elsewhere.[25] Rats were fasted for 23 h before the onset of behavioral training and refed again for 1 h later. Rats were then habituated for 5 successive days to the maze and the food reward (5 min of exposure to the maze each day, with food pellets initially scattered along the floor then food was gradually restricted to both arms of the maze) throughout drug treatment.[26,27]

The T maze procedure was modified based on the results of a pilot study, where each rat received throughout drug treatment 2 daily alternation sessions, incremented to 4 sessions and up to 6 daily sessions thereafter. Then rats were trained for 3 days and received 6 sessions daily. Each session consisted of 2 trials with 30-s

delay between the first and second trials. On the first trial the rat was rewarded with a food pellet for entry into either of the choice arms. After eating the pellet the rat was returned back to the holding box. The choice points and the entire maze were wiped with alcohol solution between trials. On the second trial, a reward was placed only in the choice arm opposite to that visited in the first trial. The intertrial interval was approximately 2 min.

A correct choice was defined as the rat alternated the arm (entered its four paws into the choice arm opposite to that entered in the first trial). The T maze alternation task was also assessed 15 min following scopolamine injection.

$$\%\ \text{of basal correct trials} = [\text{number of correct trials after scopolamine treatment/baseline of correct trials(mean of 3 days training throughout drug treatment)}] \times 100.$$

$$\%\ \text{of correct trials(\% of control)} = (\text{square root transformed \% of basal correct trials of normal or treated groups/square root transformed \% of basal correct trials of control group}) \times 100.$$

Square root transformed percent was calculated according to previous method.[28]

#### 2.4.2. Object recognition test

The test apparatus was designed as described elsewhere.[29] Three days before testing, each rat was allowed to explore the apparatus for 2 min, while on the testing day, 30 min following scopolamine injection, a session of two trials, 2-min each was allowed. In the "sample" trial (T1), two identical objects were placed in two opposite corners of the apparatus. A rat was placed in the apparatus and was left to explore these two identical objects. After T1, the rat was placed back in its home cage and an inter-trial interval of 1 h was given. Subsequently, the "choice" trial (T2) was performed. In T2, a new object (N) replaced one of the objects that were presented in T1, then rats were exposed again to two different objects: the familiar (F) and the new one (N).

Exploration was defined as follows: directing the nose toward the object at a distance of no more than 2 cm and/or touching the object with the nose.

From this measure, a series of variables were then calculated: the total time spent in exploring the two identical objects in T1, and that spent in exploring the two different objects, F and N in T2. The discrimination between F and N in T2 was measured by comparing the time spent in exploring the F with that spent in exploring the N. DI is the discrimination index and represents the difference in exploration time expressed as a proportion of the total time spent exploring the two objects in T2. DI was then calculated; $DI = N - F/N + F$.

### 2.5. Brain homogenate preparation

Rats were sacrificed by decapitation immediately after object recognition test. The whole brain was carefully excised, immediately weighed to avoid drying and stored at −80 °C. Brain was homogenized in ice-cold phosphate buffer (20% w/v). The homogenate was used for determination of brain contents of MDA, GSH, brain cholinesterase activity and TNF-content.

### 2.6. Determination of biochemical markers in brain homogenate

Brain MDA content was determined according to the method of Ruiz-Larrea et al.[30] the supernatant was read spectrophotometrically at 532 nm and MDA brain content was expressed as nmol/g tissue. The brain GSH content was determined according to the methods described elsewhere.[31,32] Calculation of GSH content was based on a standard glutathione curve and expressed as μmol/g tissue. Brain cholinesterase activity was determined colorimetrically using Quimica Clinica Aplicada S.A. kit according to the method described previously.[33] The mean of absorbance change per 30 s (ΔA/30 s) was determined for every reading. Cholinesterase activity was expressed as U/g tissue. TNF-α was estimated using rat specific immunoassay kit (RayBio® Rat TNF-alpha ELISA) according to the method described previously.[34] The intensity of the colored product is directly proportional to the concentration of rat TNF-α using a microplate reader set at 450 nm and expressed as ng/g tissue. The sample values are then read off the standard curve.

### 2.7. Statistical analysis

Data concerning the T maze alternation task, object recognition test and biochemical parameters were expressed as mean ± SEM. Comparison between more than two groups was carried out using one-way analysis of variance (ANOVA) followed by Tukey's multiple comparison test. Comparison with baseline in the T maze alternation task was carried out by two-way ANOVA followed by Bonferroni multiple comparison test. When comparing within each group the exploration times of (T1 vs. T2) in the object recognition test and the exploration times of the F and N objects in T2, Student's *t*-test was used. All analyses utilized Graph pad Prism 5.0 statistical package for Windows (Graphpad Software Inc., San Diego, USA). Statistical significance was set at $P < 0.05$.

## 3. Results

### 3.1. Effect of NSO or WGO on scopolamine-induced memory impairment in the T maze alternation task

Treatment of rats with scopolamine significantly reduced correct trials to be 44% of their basal correct choices at 30-s interval delay and significantly reduced correct trials to be 67% of that in normal rats (Table 1 and Fig. 1).

Daily oral administration of NSO (0.92 mg/kg) or WGO (170 mg/kg) for 14 consecutive days before scopolamine significantly increased the correct trials to be 142% and 147% of that in the control scopolamine group, respectively to be similar to that of the donepezil group (Fig. 1).

### 3.2. Effect of NSO or WGO on scopolamine-induced memory impairment in the object recognition test

Within T1 and T2, no differences in total exploration time were seen among the different groups. Comparing with the

**Table 1** Influence of 14 daily administration of NSO or WGO on performance of scopolamine-treated rats in the T maze alternation task.

| Treatment | Parameters | |
|---|---|---|
| | Spatial working memory | |
| | Correct trials | |
| | Baseline (mean of 3 days training) throughout drug treatment | After scopolamine treatment |
| Normal (saline) | 4.83[@] ± 0.26 | 4.70[@] ± 0.30 |
| Control (Scopolamine) (16 mg/kg, i.p.) | 4.73[@] ± 0.27 | 2.10[*] ± 0.23 |
| Donepezil (10 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 5.47[@] ± 0.12 | 5.00[@] ± 0.30 |
| NSO (0.92 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 5.17[@] ± 0.20 | 4.60[@] ± 0.34 |
| WGO (170 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 5.23[@] ± 0.22 | 4.90[@] ± 0.28 |

Data represent means ± SEM ($n = 10$). Statistical analysis was carried out by two-way ANOVA followed by Bonferroni multiple comparison test.
[*]Significant difference from baseline at $P < 0.05$.
[@]Significant difference from correct trials after scopolamine treatment at $P < 0.05$.

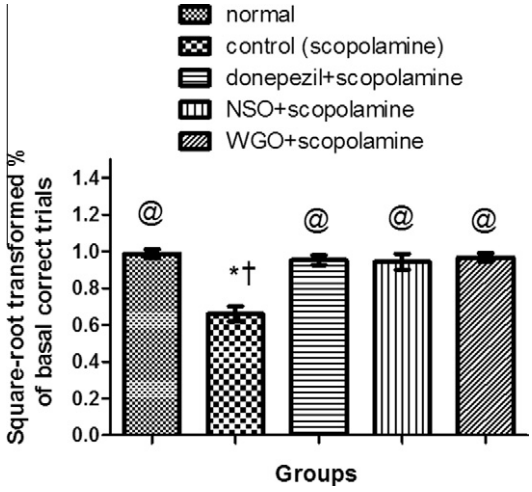


**Figure 1** Effect of NSO or WGO on scopolamine-induced memory impairment in the T maze alternation task. Rats received 14 daily administration of NSO (0.92 mg/kg, p.o.), WGO (170 mg/kg, p.o.) or donepezil (10 mg/kg, p.o.) used as a standard drug. The normal and control groups similarly received saline. Scopolamine was administered as a single dose (16 mg/kg, i.p.) 30 min after the 14th treatment of all groups except the normal one. Fifteen minutes later the task was carried out. Data are expressed as square- root transformed % of basal correct trials ($n = 10$). Statistical analysis was carried out by one-way ANOVA followed by Tukey's multiple comparison test.[*]Significant difference from normal group at $p < 0.05$. [@]Significant difference from control (scopolamine) group at $P < 0.05$. [†]Significant difference from donepezil group at $p < 0.05$.

respective exploratory activity exhibited in T1, a reduction of it in T2 was observed in all rats except those treated with scopolamine (Fig. 2). Rats treated with scopolamine explored the novel and familiar objects similarly. Exploration time in T2 showed that NSO or WGO-treated rats were similar to donepezil-treated rats and explored the new object significantly more than the familiar one as shown in Fig. 3. DI revealed that all rats, except those treated with scopolamine, significantly discriminated the new object better than the familiar one (Fig. 4).

### 3.3. Malondialdehyde brain content

The MDA content expressed as nmol/g tissue was significantly higher in scopolamine-treated rats 164.01 ± 1.54 as compared to the normal group 150.13 ± 2.92. NSO administered for 14 successive days before scopolamine resulted in a significant decrease in MDA content to 87% of scopolamine control group and restored MDA brain content to be similar to donepezil. WGO did not show any change in MDA content when compared to normal, scopolamine and donepezil groups (Table 2).

### 3.4. Glutathione brain content

A significant decrease in GSH content was observed in the scopolamine-treated group 1.57 ± 0.03 as compared to the normal group 2.45 ± 0.09 expressed as μmol/g tissue. NSO and WGO administered for 14 successive days before scopolamine showed a significant increase in GSH brain content to 142% and 160% of scopolamine control group, respectively. NSO and WGO increased brain GSH content to a similar level as the donepezil group (Table 2).

### 3.5. Brain cholinesterase activity

There was a significant increase in cholinesterase activity in the scopolamine-treated group 475.72 ± 40.90 as compared to the normal group 355.16 ± 28.74, expressed as U/g tissue. NSO did not alter cholinesterase activity when compared to the scopolamine and donepezil groups. WGO administered before scopolamine resulted in a significant decrease in cholinesterase activity to 72% of scopolamine control group (Table 3).

### 3.6. Tumor necrosis factor-alpha brain content

Scopolamine administered as a single intraperitoneal dose (16 mg/kg) significantly increased TNF-α content to be 7.07 ± 0.56 whereas the normal group was 2.68 ± 0.07, expressed as ng/g tissue. NSO administered for 14 successive days before scopolamine resulted in a significant decrease in TNF-α content to 26% of scopolamine control group. NSO decreased the

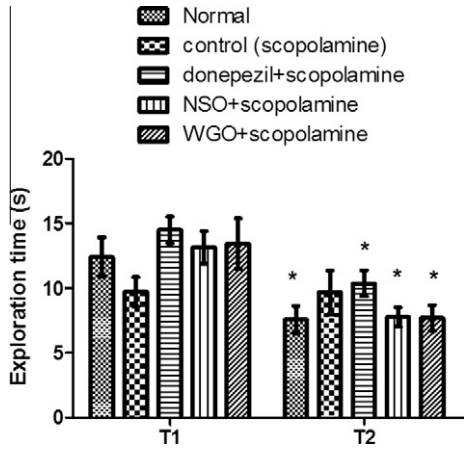


**Figure 2** Effect of NSO or WGO on exploration time in T1 and T2 in scopolamine-induced memory impairment in rats using the object recognition test. Rats received 14 daily administration of NSO (0.92 mg/kg, p.o.), WGO (170 mg/kg, p.o.) or donepezil (10 mg/kg, p.o.) used as a standard drug. The normal and control groups similarly received saline. Scopolamine was administered as a single dose (16 mg/kg, i.p.) 30 min after the 14th treatment of all groups except the normal one. T1 was performed 30 min following scopolamine. T2 was carried out 60 min following T1. Results are expressed as mean ± SEM ($n = 8$). Statistical analysis was carried out by Student's *t*-test. *Significant difference from the corresponding T1 group at $P < 0.05$.

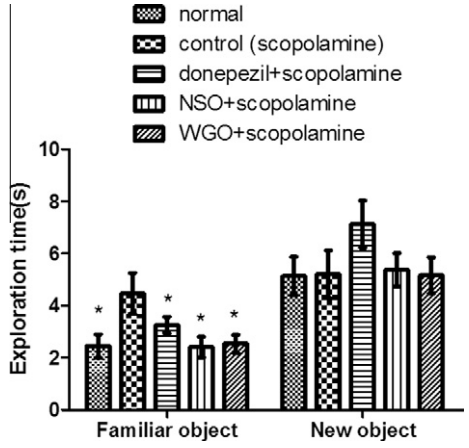


**Figure 3** Effect of NSO or WGO on the exploration time of the familiar (F) vs. the novel object (N) in T2 in the object recognition test. Rats received 14 daily administration of NSO (0.92 mg/kg, p.o.), WGO (170 mg/kg, p.o.) or donepezil (10 mg/kg, p.o.) used as a standard drug. The normal and control groups similarly received saline. Scopolamine was administered as a single dose (16 mg/kg, i.p.) 30 min after the 14th treatment of all groups except the normal one. T1 was performed 30 min following scopolamine. T2 was carried out 60 min following T1. Data represent mean ± SEM ($n = 8$). Statistical analysis was carried out by Student's *t*-test. *Significant difference from the corresponding N group at $P < 0.05$.

TNF-α content to a similar level as the donepezil group. WGO did not show any change in TNF-α content when compared to

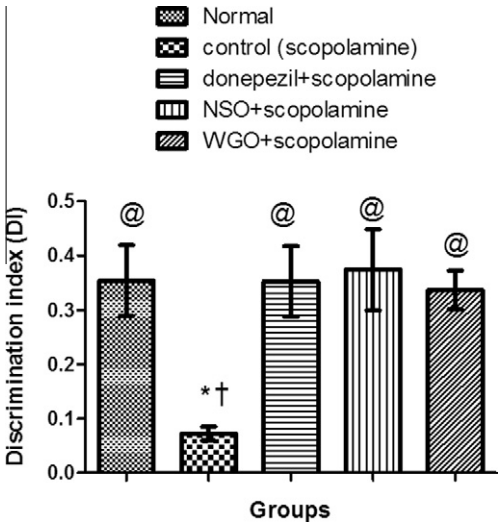


**Figure 4** Effect of NSO or WGO on discrimination index (DI) of scopolamine-induced memory impairment in rats using the object recognition test. Rats received 14 daily administration of NSO (0.92 mg/kg, p.o.), WGO (170 mg/kg, p.o.) or donepezil (10 mg/kg, p.o.) used as a standard drug. The normal and control groups similarly received saline. Scopolamine was administered as a single dose (16 mg/kg, i.p.) 30 min after the 14th treatment of all groups except the normal one. T1 was performed 30 min following scopolamine. T2 was carried out 60 min following T1. Data represent mean ± SEM ($n = 8$). Statistical analysis was carried out by one-way ANOVA followed by Tukey's multiple comparison test. *Significant difference from normal group at $p < 0.05$. @Significant difference from control (scopolamine) group at $P < 0.05$. † Significant difference from donepezil group at $p < 0.05$.

control scopolamine group and significantly elevated TNF-α content when compared to donepezil group (Table 4).

## 4. Discussion

NSO in this study significantly antagonized scopolamine-induced cognitive dysfunction in the T maze alternation task. NSO ameliorating effects on rats' performance in the T maze alternation task were similar to donepezil, indicating that NSO attenuated the short-term memory deficit induced by scopolamine in rats.

Similarly, in the current study WGO significantly reversed scopolamine-induced cognitive dysfunction in the T maze alternation task and enhanced rats' performance to be similar to donepezil and this indicate that WGO restored the spatial working memory in demented rats. As WGO is the natural supplement of vitamin E, so our result is in agreement with another study which stated that vitamin E attenuated scopolamine's effect on memory retention of passive avoidance learning in rats.[35] Moreover, vitamin E ameliorated memory impairments during the normal aging process in both humans and animals.[36,37]

The current object recognition paradigm revealed that no differences were observed among the different populations within T1 and T2, while the overall exploratory activity of all experimental groups in T2 was reduced as compared to their performance in T1 except in scopolamine treated rats. This

**Table 2** Effect of NSO or WGO on brain MDA and GSH contents in scopolamine-treated rats.

| Groups | Parameters | |
|---|---|---|
| | MDA (nmol/g tissue) | GSH (*u*mol/g tissue) |
| Normal (saline) | 150.13[@] ± 2.92 | 2.45[@] ± 0.09 |
| Control (Scopolamine) (16 mg/kg, i.p.) | 164.01[*,†] ± 1.54 | 1.57[*,†] ± 0.03 |
| Donepezil (10 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 148.40[@] ± 1.88 | 2.78[@] ± 0.21 |
| NSO (0.92 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 143.21[@] ± 4.35 | 2.23[@] ± 0.21 |
| WGO (170 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 154.90 ± 3.52 | 2.51[@] ± 0.15 |

Data represent means ± SEM ($n = 6$). Statistical analysis was carried out by one-way ANOVA followed by Tukey's multiple comparison test.
[*]Significant difference from normal group at $P < 0.05$.
[@]Significant difference from control (scopolamine) group at $P < 0.05$.
[†]Significant difference from donepezil group at $P < 0.05$.

**Table 3** Effect of NSO or WGO on brain cholinesterase activity of scopolamine-treated rats.

| Groups | Parameters Cholinesterase activity (U/g tissue) |
|---|---|
| Normal (saline) | 355.16[@] ± 28.74 |
| Control (Scopolamine) (16 mg/kg, i.p.) | 475.72[*] ± 40.90 |
| Donepezil (10 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 379.27 ± 6.39 |
| NSO (0.92 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 420.33 ± 18.72 |
| WGO (170 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 344.08[@] ± 30.95 |

Data represent means ± SEM ($n = 6$). Statistical analysis was carried out by one-way ANOVA followed by Tukey's multiple comparison test.
[*]Significant difference from normal group at $P < 0.05$.
[@] Significant difference from control (scopolamine) group at $P < 0.05$.

**Table 4** Effect of NSO or WGO on TNF-α content of scopolamine-treated rats.

| Groups | Parameters TNF-α (ng/g tissue) |
|---|---|
| Normal (saline) | 2.68[@] ± 0.07 |
| Control (Scopolamine) (16 mg/kg, i.p.) | 7.07[*,†] ± 0.56 |
| Donepezil (10 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 2.78[@] ± 0.22 |
| NSO (0.92 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 1.86[@] ± 0.15 |
| WGO (170 mg/kg, p.o.) + Scopolamine (16 mg/kg, i.p.) | 7.9[*,†] ± 0.61 |

Data represent means ± SEM ($n = 6$). Statistical analysis was carried out by one-way ANOVA followed by Tukey's multiple comparison test.
[*]Significant difference from normal group at $P < 0.05$.
[@]Significant difference from control (scopolamine) group at $P < 0.05$.
[†]Significant difference from donepezil group at $P < 0.05$.

observation is supported by previous studies that as scopolamine is delivered systemically, therefore it cannot be excluded that non specific factors (attentional, sensorimotor) might have influenced rats' performance.[38] This also implies that the effects of NSO and WGO on cognitive performance did not interfere with sensorimotor performance of rats. NSO or WGO-treated rats were similar to donepezil-treated rats and completely reversed the object recognition dysfunction induced by scopolamine, by reducing the time spent in exploring the familiar object and therefore were able to discriminate between the familiar and new object, indicating that NSO and WGO improved non-spatial working memory deficit in amnesic rats.

Several studies suggest that oxidative stress plays an important role in pathogenesis of neurodegenerative disorders like AD.[39] Thus, the progression of neurodegenerative disorder can be inhibited by the use of free radical scavengers and anti-oxidants. MDA is an end product of lipid peroxidation; a measure of free radical generation. GSH is an essential tripeptide, an antioxidant found in all animal cells. It reacts with the free radicals and can protect cells from singlet oxygen, hydroxyl radical and superoxide radical damage.[40,41]

Pretreatment of scopolamine memory deficit rats with NSO resulted in a significant decrease in MDA and increase in GSH brain contents to be similar to that observed in the donepezil group. The present findings imply the neuroprotective action of NSO through its significant antioxidant activity against scopolamine-induced oxidative stress. NSO, known as an antioxidant agent, ameliorated oxidative injury in the tissues and functional deteriorations.[42] The maintenance of normal GSH level was also reported to be important for acquisition of spatial memory since GSH unavailability induced failures in hippocampal synaptic plasticity mechanisms that were related to spatial memory deficits.[43]

WGO resulted in a significant increase in brain GSH content to be similar to the donepezil group. This finding is in harmony with previous results which reported that vitamin E is a major antioxidant in biological systems[44] that can prevent or decrease the harmful effects of oxidative stress in different tissues.[45,46] The current finding supports the notion that antioxidants are considered to be a promising approach to neuroprotection against AD.[47]

Administration of donepezil prior to scopolamine in this study did not alter cholinesterase activity as compared to scopolamine control group. This finding is in conformity with previous study,[48] confirming that selective AChEIs do not alter BChE activity.

NSO treated scopolamine group did not show any change in cholinesterase activity when compared with scopolamine

and donepezil groups. However, it still shared comparable memory enhancing effects in the T maze alternation task and object recognition test, suggesting that NSO enhances memory in this model possibly through mechanism(s) independent of the cholinesterase enzyme inhibition as its neuroprotective effects against beta amyloid (Aβ) toxicity may play a potential role in preventing AD progression.[49]

The results of WGO treated scopolamine group showed significant decrease in cholinesterase activity as compared to control scopolamine group. This study is in agreement with previous findings which demonstrated that rats pretreated with vitamins E and C prevented Arginine induced alteration of BChE activity in serum of rats.[50] Inhibition of BChE may delay plaque formation and enhance the magnitude of long term potentiation,[51] thus suggesting the possibility to use WGO as butyrylcholinesterase inhibitor (BChEI) in the treatment of AD not only to increase the availability of acetylcholine at the synapses, but also to reduce the number of plaques and increase synaptic plasticity.

TNF-α has been postulated to be an important mediating factor in the neurodegeneration seen in AD[52] and may contribute to cognitive dysfunction and accelerated progression of AD.[53]

Treatment with NSO before scopolamine significantly decreased TNF-α brain content to a level similar to that observed in the donepezil group. Thymoquinone (TQ) the main active principle in NSO has been reported to be effective against transient forebrain ischemia-induced inflammation in the rat hippocampus.[54] In this study, it is likely that scopolamine-induced oxidative stress may lead to inflammation. These findings imply that NSO mitigated scopolamine-induced memory impairment through its anti-inflammatory action probably via an anti-oxidative mechanism.

WGO resulted in a significant increase in TNF-α content when compared to that of donepezil group. This finding suggests that WGO cognitive enhancing effect in this model may be possibly via mechanism(s) independent on inhibition of elevated TNF-α brain content as it was reported that vitamin E supplementation reduced Aβ deposition in a young transgenic mouse model of AD (Tg2576).[55]

In conclusion, both NSO and WGO exerted cognitive enhancing effects against scopolamine-induced memory impairment in rats. Therefore, their use might offer a useful therapeutic choice in either prevention or treatment of AD.

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