J. Phytomol. Pharmacol., 3(1), 01-07, 2024
https://doi.org/10.56717/jpp.2024.v03i01.022
ISSN(Print): 2957-5818, ISSN(Online): 2958-6224

Journal of Phytomolecules & Pharmacology

Research Article

Antidepressant activity of fresh young shoot essential oil of *Asparagus officinalis* L. in mice

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Abstract

Asparagus officinalis L. is used to treat nervous disorders but there is a lack of scientific investigation to validate this ethnomedicinal claim. Essential oils have been reported to demonstrate central nervous system activities including antidepressant activity. Hence, this study investigated the antidepressant effect of the essential oil of *Asparagus officinalis* L. (EOAO). The antidepressant effect of EOAO was investigated using forced swimming test (FST) and tail suspension test (TST) in mice. The possible neural mechanism of its antidepressant effect was investigated using cyproheptadine, yohimbine, atropine and haloperidol on the tail suspension model. The EOAO significantly ($p<0.05$-0.001) decreased the immobility time of mice in FST and TST showing antidepressant effect. This effect was significantly ($p<0.01$-0.001) reversed by yohimbine (1 mg/kg, i.p.), atropine (1 mg/kg, i.p.), haloperidol (0.25 mg/kg, i.p), suggesting the involvement of adrenergic, cholinergic and dopaminergic signaling pathways. This study shows that essential oil of *A. officinalis* L. demonstrated significant antidepressant activity on the two models used and that the mechanism of its action could probably be through adrenergic, cholinergic and dopaminergic pathways.

Article Information
Received: 10 January 2024
Revised: 01 February 2024
Accepted: 01 February 2024
Published: 01 March 2024

Academic Editor
Prof. Dr. Marcello Iriti

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Keywords
Asparagus officinalis L., Asparagaceae, essential oil, antidepressant activity, forced swimming test, tail suspension test, mechanism

1. Introduction

Depression is a common mental disorder seen in about 5% of the global adult population. It is an underlying cause of suicide and more women are affected than men [1]. Neurotransmitters (norepinephrine, serotonin, dopamine, glutamate), nitric oxide, a second messenger are implicated in the pathophysiology and treatment of depression [2]. The remedial approach to depression includes psychological treatments for mild depression and the use of medications for moderate to severe depression [3]. Despite the available treatment, over 75% of people living with depression in low- and middle-income countries do not access treatment [3]. Antidepressant medications belonging to different classes (tricyclic antidepressants, selective serotonin reuptake inhibitors, monoamine oxidase inhibitors, 5-HT_2 receptor antagonists, specific serotonin-norepinephrine reuptake inhibitors, and other heterocyclics) are clinically employed as drug therapy for depression. However, they are limited by side effects, some of which include sexual dysfunction, apathy, fatigue and cognitive impairment [2]. Herbs have been used to treat various disorders as a form of alternative medicine [4] and this is a practice in many cultures [5]. The healthcare needs of about 80% of the world population largely depend on medicines from plant sources [6] and several plant species have been evaluated to resolve various ailments [7]. The challenge of cost, availability, side effects and adulteration of synthetic drugs in developing countries is paving the way for the search of new drugs that are plant-based [8]. Asparagus is cultivated

as a vegetable and medicinal herb. In ethnomedicine, the roots and the shoots are of medicinal importance. The Asparagus genus, comprising over 300 species is widely evaluated for their nutritional, biological and pharmacological actions. Asparagus species are naturally cultivated in Asia, Africa and Europe [9]. In traditional medicine edible herbs such as Asparagus have widely become important ingredients in functional foods and drugs. The roots of *Asparagus officinalis* L. are used to promote improvement in physical and mental health [10-12]. Aqueous extracts from *Asparagus officinalis* L. stems in a study protected learning and memory function in mice [13]. Essential oils have been reported to demonstrate central nervous system activities including antidepressant activity [14] in different animal models. This study was aimed at evaluating the potential antidepressant effect of the essential oil of the fresh young shoot of *Asparagus officinalis* L. as well as its probable neural mechanism(s) of action and provide evidence-based reports on the use of aromatherapy in treating depression.

2. Materials and methods

2.1 Plant material and extraction of essential oil

2.1.1 Plant collection and identification

The plant, *A. officinalis* L. was collected at Kajola-Ajile in Ile-Ife, Nigeria. It was identified and authenticated by Mr I.I. Ogunlowo, the herbarium officer of the Pharmacognosy Department, Faculty of Pharmacy, Obafemi Awolowo University, Ile-Ife and herbarium voucher number FPI12348 was issued.

2.1.2 Extraction of essential oil

The volatile fraction of *A. officinalis* L. fresh young shoot was obtained by a process of hydro–distillation for about 4 h using Clevenger-type apparatus, at the Postgraduate Toxicology Laboratory, Department of Biochemistry, Faculty of Science, Obafemi Awolowo University, Ile – Ife, Nigeria. Pale yellow oil produced was collected after cooling and stored in the refrigerator (4°C) before use.

2.2 Antidepressant activity assay

2.2.1 Preparation of test oil

The Essential oil of *A. officinalis* L. (EOAO) was weighed and emulsified with Tween - 80 and diluted with distilled water to the required concentration.

2.2.2 Drugs and treatments

The following drugs were used as either standard or receptor antagonists in the study: imipramine (Sigma, Switzerland, MSDS), diazepam (Roche), yohimbine hydrochloride, atropine sulphate, cyproheptadine hydrochloride and haloperidol.

2.2.3 Laboratory animals

Swiss mice (18 - 22 g) were purchased from the Animal House of the Faculty of Pharmacy, OAU, Ile-Ife, Osun State, Nigeria. The mice were kept in cages and fed on standard animal pellets and water *ad libitum*. The experimental protocols adopted in these investigations complied with the approved institutional guidelines which align with the internationally accepted principles for Laboratory Animal Use and Care [15].

2.2.4 Forced swimming test (FST)

The test was carried out as described by Akinpelu et al. [16]. Mice were placed individually into cylindrical chambers (height 30 cm, diameter 20 cm) filled up to the 15 cm mark with water maintained at 25°C. Mice were placed in the cylinder for 6 min. Mobility and immobility were assessed during the last 4 min of the total duration of the test (6 min). The parameter scored for mobility was struggling which consists of swimming (when the animal paddles with its limbs) or climbing (when the mouse makes an attempt to jump out of the water or uses its forelimbs to climb the wall of the container). A mouse was judged immobile when it passively floats just to keep its head above water.

2.2.5 Tail suspension test (TST)

The TST was performed to assess the antidepressant activity [17]. 30 min following administration of *A. officinalis* L., mice were suspended by the tail on the edge of a platform, 30 cm above a table surface, with the aid of an adhesive tape placed approximately 1 cm from the tip of the tail. The animal was suspended for 5 min, and the duration of immobility was assessed. The total time mice hung passively and were completely motionless was estimated as the immobility time. Imipramine (10 mg/kg, i.p.) was used as the reference drug.

2.2.6 Exploratory activity in the open-field test (OFT)

The spontaneous locomotor and exploratory activities were assessed in an OFT as described previously by Rogoz et al. [18]. 30 min after intraperitoneal treatments with the vehicle, *A. officinalis* L. and 30 min

https://doi.org/10.56717/jpp.2024.v03i01.022

after injection of diazepam (1 mg/kg, i.p). Each mouse was gently placed in an observation cage (100 cm x 100 cm x 30 cm) subdivided into 16 equal squares on its floor. The number of squares crossed and rearing made for 5 min duration was scored.

2.2.7 Mechanism of the antidepressant effect of A. officinalis L.

A. officinalis L. (25 mg/kg, i.p.) was used for the determination of the probable neural mechanism of action. Mice were pretreated with adrenergic (α_2) receptor blocker, yohimbine (1 mg/kg, i.p.), 5-HT_2 serotonergic receptor blocker [cyproheptadine (2 mg/kg, i.p.)], dopaminergic receptor blocker [haloperidol (0.25 mg/kg, i.p., D_2 receptor blocker)], cholinergic receptor blocker [atropine (1 mg/kg, i.p) 30 min before EOAO (25 mg/kg, i.p.). Thirty minutes after intraperitoneal administration of EOAO, the mice were subjected to TST.

2.3 Statistical analysis

All data are presented as mean ± SEM and analysis was done using one-way analysis of variance (ANOVA) followed by post-hoc test using Dunnett's comparison test. $P<0.05$ was considered statistically significant.

3. Results and Discussion

The results showed evidence of the potential antidepressant properties of the essential oil of *Asparagus officinalis* L. in mice. The essential oil of *Asparagus officinalis* L. significantly increased the mobility time of mice in both the FST and the TST models compared to the vehicle (Figs 1, 2 A and B). The immobility phase is termed behavioural despair in animals and this is similar to depression in humans [19]. Imipramine (tricyclic antidepressant) and fluoxetine (selective serotonin reuptake inhibitor) have been reported to give a similar effect of reversing immobility posture and promoting escape-related behaviour as also observed for essential oil of *Asparagus officinalis* L. in mice on the FST and TST models [20-22]. Increased mobility time in mice on FST and TST is similar to increased clinical effects of antidepressant drugs [23]. Studies have also shown that agents that reduce the immobility time on FST and TST models possess antidepressant-like properties [24-25].

TST and FST are commonly used to screen potential

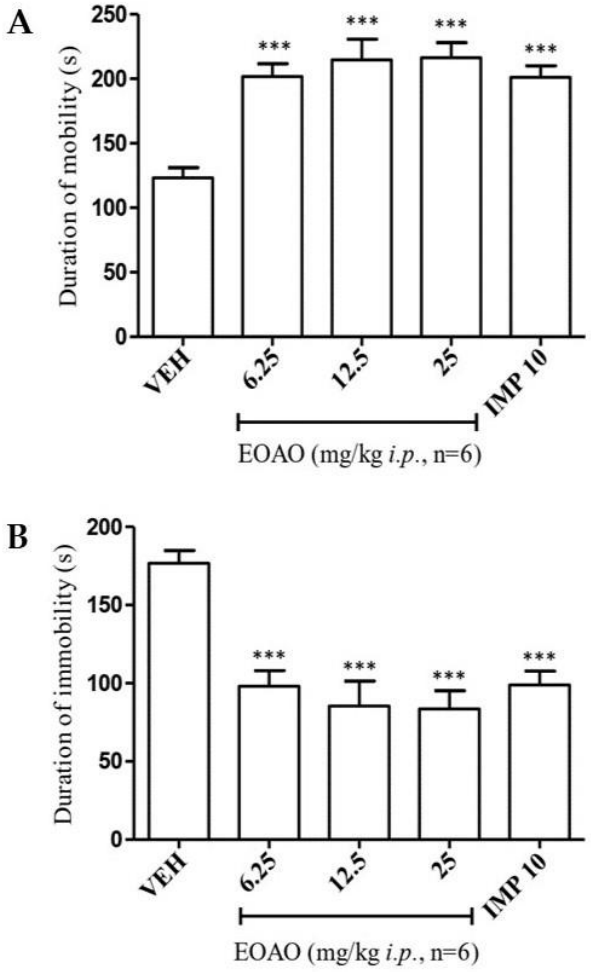

Figure 1. A and B: Effect of essential oil of *Asparagus officinalis* L. on duration of mobility (A) and duration of immobility (B) in tail suspension test.
[VEH, EOAO and IMP represent vehicle (5% Tween 80), essential oil of *Asparagus officinalis* L. and imipramine respectively.
***P < 0.001, **p<0.01, *p<0.05 statistically significant compared to the vehicle (ANOVA, Dunnett's test)].

antidepressant drugs. Tricyclics, serotonin-specific reuptake inhibitors, monoamine oxidase inhibitors, and atypicals are classes of antidepressants that have been assessed using the TST and FST [19-20]. Previous studies have shown that agents that cause alteration in the locomotor activity of mice may result in false positive/negative results on both FST and TST models [26]. To ascertain the antidepressant properties of the essential oil, its effect in mice in the Open Field Test (OFT) was investigated. The OFT is used to evaluate locomotor and exploratory activities in rodents [27]. The result showed that the antidepressant effect of the essential oil of *A. officinalis* L. was not due to psychostimulant action, as there was no significant difference in rearing and locomotion in mice across

https://doi.org/10.56717/jpp.2024.v03i01.022

Table 1. Effects of EOAO and diazepam on behaviour of mice in Open field paradigm

Group	Dose (mg/kg, i.p.)	Frequency of rearing/5 min	Frequency of locomotion/ 5 min
5% Tween 80	0.1 ml/10 g	43.00 ± 3.85	83.17 ± 5.92
EOAO	6.25	40.67 ± 2.43	105.2 ± 9.58
	12.5	35.17 ± 4.52	81.17 ± 3.90
	25	40.00 ± 1.29	88.17 ± 2.46
Diazepam	1	32.00 ± 2.79	83.00 ± 8.44

the tested groups when compared to the vehicle. This further reinforces that the anti-depressant property observed is not false positive (Table 1).

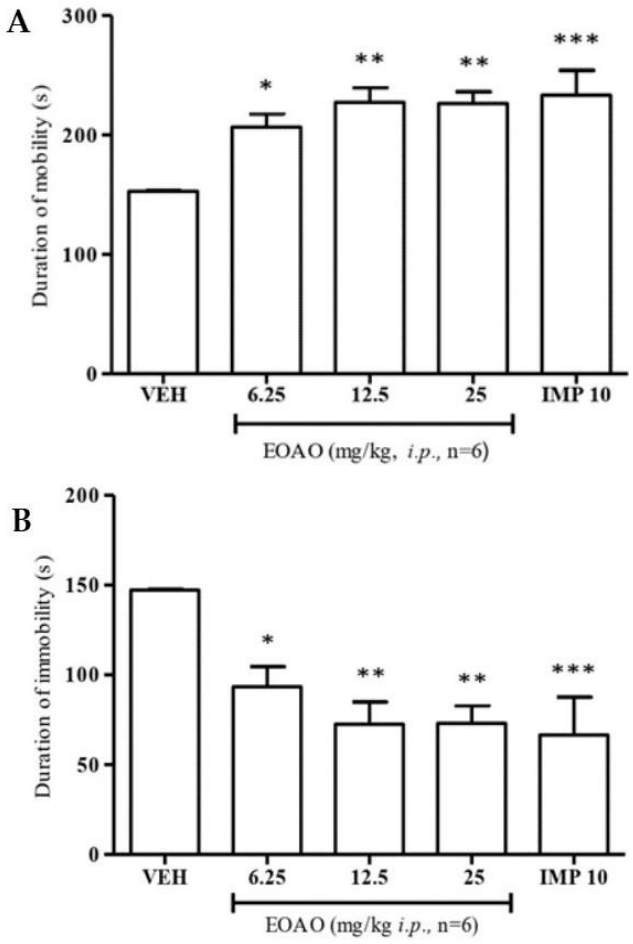

Figure 2. A and B: Effect of essential oil of *Asparagus officinalis* L. on duration of mobility (A) and duration of immobility (B) in forced swimming test.

VEH, EOAO and IMP represent vehicle (5% Tween 80), essential oil of *Asparagus officinalis* L. and imipramine respectively. ***P < 0.001, **p<0.01, *p<0.05 statistically significant compared to the vehicle (ANOVA, Dunnett's test).

Several studies have reported the efficiency of essential oils in relieving symptoms of diseases related to mental disorders, improving mood and mental well-being, with the advantage of fewer adverse reactions [28]. Some of the popularly consumed essential oils used in relieving symptoms of depression and anxiety amongst others are from rose (*Rosa damascena* Mill.), patchouli (*Pogostemon cablin* (Blanco) Benth.), lemongrass (*Cymbopogon citratus* (DC.) Stapf), sandalwood (*Santalum album* L.), bergamot (*Citrus bergamia* Risso & Poit.), valerian (*Valeriana officinalis* L.) and lemon (*Citrus limonum* Risso) [29-31]. In clinical trials, Lavender (*Lavandula angustifolia* Mill.) essential oil has been studied and shown to alleviate symptoms of anxiety and depression [32] and as a positive control in an antidepressant study [33].

The probable neural mechanism of action of *A. officinalis* L. essential oil was evaluated in this study using adrenergic (α_2) receptor blocker, yohimbine (1 mg/kg, i.p.), 5-HT_2 serotonergic receptor blocker [cyproheptadine (2 mg/kg, i.p.)], dopaminergic receptor blocker [haloperidol (0.25 mg/kg, i.p., D_2 receptor blocker)] and cholinergic receptor blocker [atropine (1 mg/kg, i.p)]. The results revealed a reversal of the antidepressant effect of the essential oil of *A. officinalis* L. by yohimbine, atropine and haloperidol in mice. Hence, suggesting the involvement of the adrenergic, cholinergic and dopaminergic [34-36] neurotransmission in the eliciting of its effect (Fig 3 A and B). The imipramine used as the positive control in this experiment is a typical tricyclic antidepressant used in the treatment of depression of various aetiologies. It acts by blocking the reuptake of biogenic amines (serotonin, norepinephrine, dopamine) at nerve terminals in the CNS hence, increasing postsynaptic excitation. Imipramine also acts as a muscarinic, adrenergic (α_1) and histaminergic (H_1 and H_2) receptor antagonist [37].

https://doi.org/10.56717/jpp.2024.v03i01.022

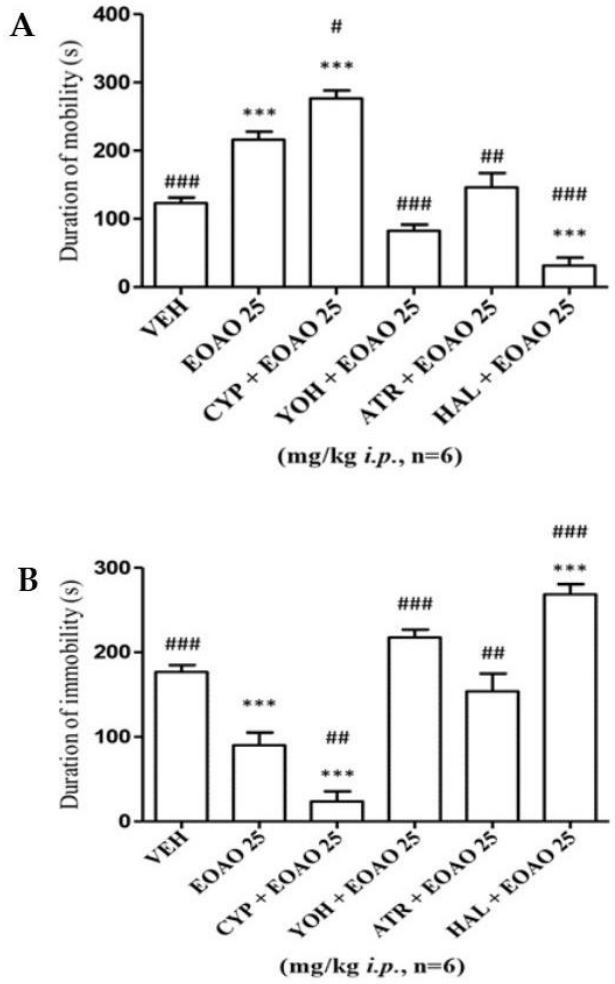

Figure 3. A and B: Effect of antagonists on essential oil of *Asparagus officinalis* L. on duration of mobility (A) and duration of immobility (B) in tail suspension test

VEH, EOAO, CYP, YOH, ATR and HAL represent vehicle (5% Tween 80), essential oil of *Asparagus officinalis* L., cyproheptadine, yohimbine, atropine and haloperidol respectively.

***P < 0.001, **p<0.01, *p<0.05 statistically significant compared to the vehicle (ANOVA, Dunnett's test).

###P < 0.001, ##p<0.01, #p<0.05 statistically significant compared to EOAO (ANOVA, Dunnett's test).

4. Conclusions

This study shows that the essential oil of *A. officinalis* L. demonstrated antidepressant activity on the two models used compared to the vehicle and that the mechanism of eliciting the antidepressant effect could probably be through adrenergic, cholinergic and dopaminergic pathways. However, further studies will be needed to identify the bioactive constituents of the oil responsible for this activity.

Authors' contributions

Conceptualization, methodology, investigation, data curation, writing and editing of the manuscript, A.S.O.; Conceptualization, supervision, review and editing of the manuscript, I.A.O.

Acknowledgements

The effort of Mr I.I. Ogunlowo in making available the plant material for hydrodistillation is appreciated.

Funding

This research received no specific grant from any funding agency (the public, commercial, or not-for-profit sectors).

Availability of data and materials

All data will be made available on request according to the journal policy.

Conflicts of interest

The authors declare no conflict of interest.

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