



Research Article

Assessing the Antidepressant Effect of Ethanol Extracts from a Combination of *Syzygium aromaticum* L. (Myrtaceae) and *Curcuma longa* L. (Zingiberaceae) on Swiss Albino Rats

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ABSTRACT

Depression is a major global health disorder associated with significant morbidity and reduced quality of life, while current pharmacotherapies are limited by adverse effects and treatment resistance. This study evaluated the Synergistic antidepressant potential of ethanol extracts of *Curcuma longa* and *Syzygium aromaticum* in albino rats. Antidepressant-like activity was assessed using the Open Field Test (OFT), Tail Suspension Test (TST), and Forced Swim Test (FST) following diazepam-induced depressive-like behavior. Animals were allocated into five groups: normal control, fluoxetine-treated positive control, and three extract-treated groups (50, 100, and 200 mg/kg; 1:1 combination). The extract exhibited dose-dependent antidepressant effects, with the 200 mg/kg dose significantly ($p < 0.01$) reducing immobility time in both TST and FST, comparable to fluoxetine. Enhanced exploratory activity in the OFT further suggested anxiolytic properties. These effects may be attributed to modulation of neurotransmitter systems, antioxidant activity, and anti-inflammatory mechanisms. The findings provide preliminary evidence supporting the antidepressant potential of *C. longa* and *S. aromaticum*, highlighting their promise as safer, plant-based alternatives or adjuncts to conventional antidepressants, warranting further mechanistic and clinical investigations.

Keywords: Antidepressant; Clove; Depression; Neuroprotection; Oxidative stress; Turmeric

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INTRODUCTION

Depression is a neuropsychiatric disorder that is characterized by prolonged feeling of sadness and reduced interest in worldly affairs that was once enjoyed. It can also impair cognitive performance such as thinking and memory, as well as appetite and sleeping pattern (World Health Organization [WHO] 2024)). Depression is a major mental health disorder impacting life-style of millions of people across the globe. According to current estimates, indicated that depression affected more than 280 million people

worldwide, ranking it as a primary cause of disability worldwide (Evans-Lacko *et al.*, 2018; Chen *et al.*, 2025).

Approximately, about 3.8 % of the world population are suffering from depression, including 5 % of adults (4 % among men and 6 % among women), and 5.7 % of adults older than 60 years. The condition is reported to occur nearly about 50 % more frequently in women than in men (WHO 2024; Liu & Kuai, 2025). According to findings by Woody *et al.*, 2017, reported that pregnant women who have given birth have

higher chances of experiencing depression than in those women who have not given birth. However, WHO 2023, reported that depression can lead to suicide, which is the fourth leading cause of death in youth between the ages of 15-29 year olds. Globally, about 700,000 individuals die as a result of suicide every year (Ulutas & Ergin 2019).

Studies globally indicate that medical students with depression often fail to recognize their condition as treatable leading to poor help-seeking behavior (Isara *et al.*, 2022). In Nigeria, the prevalence of depression indicated that about 3.1 % individuals are living with the condition, and with a significant progress in Western sub-Saharan Africa of 124.42 %. However, the total estimate of people living with depression increased worldwide by 49.86 % (Suraj *et al.*, 2021). Diazepam Induced depression-like behaviour at chronic or high-dose by potentiating the pharmacological properties of gamma-aminobutyric acid (GABA) on different sites (Nutt & Malizia 2001). Particularly, benzodiazepines bind at an inactive site at the interface link the alpha and gamma subunits on GABA-A receptor chloride ion channels. The binding causes increased in the frequency of chloride channel opening, leading to propagating the conductance of chloride ions, paradoxical sedation, reduced motor activity, and cognitive impairment (Nutt & Malizia 2001). However, with chronic administration, diazepam produces more genuine depression-like states by impairing hippocampal neuroplasticity (Monti *et al.*, 2012), reducing neurogenesis, and disrupting stress regulation via the HPA axis, thereby affecting emotional and cognitive processing (Vargas *et al.*, 2001; Walker & Herskowitz 2020).

Despite the existing antidepressant medications, depression continue to remains a major public health problem with limited availability of effective treatment options, resistance to therapy, therapy failure, and considerable side effects and of these factors might leads to suicide in some depressed patient (Zeleg-Molik & Litwa 2025). A medicinal plant refers to any plant species whose parts, such as flowers, leaves, roots, stems, fruits or seeds are utilized directly in medicinal preparations as a medicine to treat or manage a health condition or disease (Miranda, 2021). Traditional medicine are used in community and healthcare system to maintain health, diagnose, prevent diseases and improve or treat physical and mental illness (WHO, 2023). The present work aimed to evaluate the combined antidepressant potential of *Syzygium aromaticum* and *Curcuma longa*, both of which

possess significant neuroprotective properties. This research is further aimed to bridge the gap that lack existing research on their synergistic effects, thereby aiming to contribute to the development of safer and more effective alternative treatments for depression.

MATERIAL AND METHODS

Experimental Site

All experiments were carried out in the Laboratory of the department of pharmacology and toxicology Faculty of pharmacy, University of Maiduguri Borno State. The experimental procedures were conducted in accordance with the 3R (Refine, Reduce and Replace) principles to reduce the number of animal used and minimize animal suffering. Experimental protocols were reviewed and approved by the Faculty of Pharmacy, Animal Research and Ethical Committee (AREC) with approval number UM/FP/03/032.

Equipment

Beakers, syringe (1 ml and 5 ml), weighing balance, hand gloves, cotton wool, permanent markers, methylated spirits, threads, Adhesive plaster, Fluoxetine 20 mg capsule (Accord Healthcare Limited, NAFDAC Reg. PL 20075/0051)), Diazepam 10 mg/2 ml injection (JUHEL, NAFDAC Reg. No. A4-5200), Open field Apparatus, Tail Suspension Test, and Force Swim Test.

Experimental Animals

A total number of 25 healthy albino rats of both sex, weighing about 150-200 g of (16 weeks old) were used for the study. The animals were obtained from National Veterinary Research Institute NVRI, Vom Plateaus State, they were housed in a conventional wire mesh cage in a well-ventilated, regularly sanitized room. They were fed on standard diet and were allowed free water access. Animals were acclimatized for a week in condition of 12:12 h light-dark cycle, temperature of 24°C – 26°C, and a humidity of 60 % - 65 % before commencement of the experiment.

Preparation of Plant Extracts

The Fresh rhizome of Turmeric and flower bud of Clove were purchased from Monday Market, Maiduguri Borno state. The plant materials was authenticated by a Botanist at Faculty of Pharmacy Herbarium, University of Maiduguri, a voucher specimen, (Clove UMM/FPH/MYT/004; Turmeric UMM/FPH/ZIG/003) were obtained. The Ethanol Extracts of Turmeric Rhizome and clove bud were prepared using Cold Maceration method. The Fresh Rhizome of turmeric was sliced and dried, the plants were ground into fine powder and then soaked in 95 % Ethanol individually for 72 hours. The mixture was

filtered using Muslin cloth, and the solvent was evaporated under reduced pressure to obtain the concentrated extract (Wahyulianingsih *et al.*, 2016). The percentage extraction yield was calculated as follows:

Percentage Yield for Clove

$$\text{Percentage yield (\%)} = \frac{\text{Weight of dried extract (g)}}{\text{Weight of powdered plant material (g)}} \times 100$$

$$\text{Yield} = \frac{130}{1000} \times 100 = 13\%$$

Percentage yield for Tumeric

$$\text{Percentage yield (\%)} = \frac{\text{Weight of dried extract (g)}}{\text{Weight of powdered plant material (g)}} \times 100$$

$$\text{Yield} = \frac{96}{800} \times 100 = 12\%$$

Acute Toxicity Test

Acute oral toxicity studies were performed according to OECD-423 guidelines (acute toxic class method). Female rats (n=3) were fasted for 4 h with free access to water only, the extracts (clove and turmeric) were administered orally at a limit dose of 2000 mg/kg body weight to a group of three female rats. The animal were monitored for any sign of toxicity or mortality for 48 h and considered for 14 days. The doses for the combined extracts were selected based on the results of the acute toxicity study, which showed no mortality up to 2000 mg/kg. Therefore, graded doses of 50, 100, and 200 mg/kg were chosen. The combination was prepared in a 1:1 ratio, such that each dose contained equal proportions of *Syzygium aromaticum* and *Curcuma longa* extracts (25+25, 50+50, and 100+100 mg/kg, respectively).

Induction of Depression and Animal Treatment

Depression was induced with Diazepam (JUHEL) intraperitoneally at the dose of 5 mg/kg daily for a period of 7 days (Kaur *et al.*, 2016). The doses of the extracts were 50 mg/kg, 100 mg/kg, and 200 mg/kg (Anggraeni *et al.*, 2022). Oral route of administration was used for the treatment using feeding tube for 14 days (Princewill-Ogbonna *et al.*, 2025).

The Animals were randomly divided into five groups (n=5).

Group A: Normal control (received Distilled water 2 ml/kg)

Group B: Ethanol Extract of the combination of plant (High Dose 200 mg/kg)

Group C: Ethanol Extract of the combination of the plant (Medium Dose 100 mg/kg)

Group D: Ethanol Extract of the combination of the plant (Low Dose 50 mg/kg)

Group E: Positive control (Fluoxetine Capsule 10 mg/kg) (Potdar & Kibile, 2011).

Behavioural Assessment

Behavioral assessments were conducted using the same animals following completion of the treatment period. To minimize the influence of stress from one behavioral test on subsequent tests, the assays were performed on separate days in order of increasing stressfulness. The Open Field Test was conducted first to evaluate spontaneous locomotor activity and exploratory behavior, followed by the Tail Suspension Test, while the Forced Swim Test was performed last because of its greater physical and psychological stress. A minimum interval of 24 h was allowed between consecutive behavioral tests and rat was subjected to a 5 minutes duration per each test (Crawley 2000: Amin *et al.*, 2015).

Open Field Test (OFT) Day 1

The animal is taken from its cage and placed in a novel and relatively lit arena (a wooden box 70 cm x 70 cm x 50 cm, painted white and divided into squares 30 cm x 30 cm, by peripheral and central units black strips), that is large enough for the animal to move around in. Locomotors activity was assessed by observing the rats movement in the arena, time spent at the center and number of square crossed were recorded (Fuchs and Flugge, 2006).

Tail Suspension Test (TST) Day 2

TST is a common test for despair-like behavior, a symptom of depression, in which a mouse is suspended by the tail and the amount of time the animal is in an immobile state is measured so also the mobile state (McNamara *et al.*, 2020).

Forced Swim Test (FST) Day 3

Forced Swim Test is carried out by placing a rodent in a cylinder (20 cm in diameter) filled with water (50 cm at room temperature) so that animal could not touch the bottom of the container with their tails or the hind paws and observing how much time is required for the rodent to become immobile after multiple escape attempt. It has been shown that antidepressant treatment decreases the duration of immobility exhibited by the rodents (kaufman *et al.*, 2016).

Data Analysis

The data from each group were presented as mean $\pm$ SEM (n = 5 per group) using Microsoft Excel and statistical analysis was performed using two-way repeated-measures ANOVA. Post hoc tests Turkey's HSD was conducted to identify specific group differences. A significance level of $p < 0.05$ was considered statistically significant.

RESULT

The OFT results showed a reduction in the time spent in the centre of the open field by the rats, indicating anxiety-like behaviour following induction with diazepam. However, after treatment, a significant increases in centre time were observed in the fluoxetine-treated group (254.5%, $p < 0.01$) and in the ECT-treated groups at 100 and 200 mg/kg, with increases of 141.7% ($p < 0.05$) and 113.3% ($p < 0.01$), respectively. It is noteworthy that the extract at 50 mg/kg also increased the centre time but it is not significance (Table 1).

The TST results showed an increase in immobility time in the rats following induction, suggesting depression-like behaviour. After treatment with fluoxetine, a significant reduction in immobility time

of 86.9% was observed ($p < 0.01$). Similarly, the extract at 200 mg/kg produced a significant reduction of 85.1% ($p < 0.01$), indicating potential antidepressant-like activity. However, the extract at 50 and 100 mg/kg reduced immobility time by 71.6% and 70.2%, respectively, but these changes were not statistically significant (Table 2).

The FST results showed an increase in immobility time in the rats following induction, indicating depression-like behaviour. After treatment with fluoxetine, a significant reduction in immobility time of 69.9% was observed ($p < 0.01$). Similarly, the extracts at 100 and 200 mg/kg significantly reduced immobility time by 58.2% ($p < 0.05$) and 65.9% ($p < 0.01$), respectively, suggesting potential antidepressant-like activity. The extract at 50 mg/kg also reduced immobility time, but the change was not statistically significant (Table 3).

Table 1: Time spent at the centre of Open Field Test by the rats

Groups	Baseline (Mean $\pm$ S.E)	Post-Induction (Mean $\pm$ S.E)	Post-treatment (Mean $\pm$ S.E)	p-value	%change (increase)
DW 10 mL/kg	5.8 $\pm$ 2.2	5.4 $\pm$ 1.1	4.9 $\pm$ 0.8		-9.3%
FLX 10 mg/kg	5.6 $\pm$ 1.7	2.2 $\pm$ 0.6	7.8 $\pm$ 1.4 **	$p < 0.01$	254.5%
ECT 50 mg/kg	4.0 $\pm$ 0.8	2.0 $\pm$ 0.3	5.0 $\pm$ 0.7	NS	150.0%
ECT 100 mg/kg	5.8 $\pm$ 2.1	2.4 $\pm$ 2.1	5.8 $\pm$ 1.3*	$p < 0.05$	141.7%
ECT 200 mg/kg	4.8 $\pm$ 1.6	3.0 $\pm$ 1.0	6.4 $\pm$ 1.6*	$p < 0.01$	113.3%

Keys: DW= Distilled water, FLX= Fluoxetine, ECT= Extracts of Clove and Tumeric, NS= not significance, n=5. $p < 0.05$ and * $p < 0.01$ indicate significant differences compared with the distilled-water control at the corresponding time point. NS = not significant.

Table 2: Immobility time of the rat in tail suspension test

Groups	Baseline (Mean $\pm$ S.E)	Post-Induction (Mean $\pm$ S.E)	Post-treatment (Mean $\pm$ S.E)	p-value	% change (reduction)
DW 10 mL/kg	30.2 $\pm$ 4.3	38.6 $\pm$ 5.3	38.0 $\pm$ 4.9		1.6%
FLX 10 mg/kg	28.8 $\pm$ 4.1	109.6 $\pm$ 11.4	14.4 $\pm$ 3.8**	$p < 0.01$	86.9%
ECT 50 mg/kg	27.4 $\pm$ 3.8	107.6 $\pm$ 11.6	30.6 $\pm$ 4.8	NS	71.6%
ECT 100 mg/kg	29.8 $\pm$ 3.4	117.4 $\pm$ 11.8	35.0 $\pm$ 3.6	NS	70.2%
ECT 200 mg/kg	30.6 $\pm$ 2.7	111.6 $\pm$ 13.3	16.6 $\pm$ 3.6**	$p < 0.01$	85.1%

Keys: DW= Distilled water, FLX= Fluoxetine, ECT= Extracts of Clove and Tumeric, NS= not significance, n=5. $p < 0.05$ and * $p < 0.01$ indicate significant differences compared with the distilled-water control at the corresponding time point. NS = not significant.

Table 3: Immobility of rats in Forced Swim Test

Groups	Baseline (Mean $\pm$ S.E)	PostInduction (Mean $\pm$ S.E)	Post-treatment (Mean $\pm$ S.E)	p-value	% change (reduction)
DW 10 mL/kg	21.8 $\pm$ 3.7	26.2 $\pm$ 4.9	26.8 $\pm$ 3.5		-2.3%
FLX 10 mg/kg	22.6 $\pm$ 3.7	49.8 $\pm$ 4.3	15.0 $\pm$ 2.8**	$p < 0.01$	69.9%
ECT 50 mg/kg	24.0 $\pm$ 4.5	45.4 $\pm$ 3.0	22.6 $\pm$ 3.6	NS	50.2%
ECT 100 mg/kg	25.8 $\pm$ 4.6	49.8 $\pm$ 3.9	20.8 $\pm$ 3.3*	$p < 0.05$	58.2%
ECT 200 mg/kg	24.4 $\pm$ 3.7	44.0 $\pm$ 4.1	15.0 $\pm$ 3.1**	$p < 0.01$	65.9%

Keys: DW= Distilled water, FLX= Fluoxetine, ECT= Extracts of Clove and Tumeric, NS= not significance, n=5. $p < 0.05$ and * $p < 0.01$ indicate significant differences compared with the distilled-water control at the corresponding time point. NS = not significant.

DISCUSSION

The primary use of Open Field Test (OFT) in depression is to measure baseline locomotor activity. It ensures that any reduction in immobility seen in test like the FST or TST is due to a genuine antidepressant effect rather than a false positive caused by a drug artificially stimulating general motor activity (hyperactivity) or sedating the animal. However, because anxiety frequently co-occurs with depression, the OFT is also used to evaluate anxiety-like behaviours linked to depressive phenotypes (Kraeuter *et al.*, 2019).

In the present study (Table 1), the Open Field Test (OFT) results demonstrated that anxiety-like behaviour was successfully induced in all experimental groups except the distilled water group, as evidenced by reduced time spent in the centre of the arena following induction. Untreated rats continued to exhibit anxiety-like behaviour after treatment, with a slight decline in centre exploration. In contrast, fluoxetine produced the greatest anxiolytic effect, significantly increasing centre time by 254.5% ($p < 0.01$). Similarly, the extract of clove and turmeric (ECT) improved centre exploration in a dose-dependent manner, with 100 and 200 mg/kg producing statistically significant increases ($p < 0.05$ and $p < 0.01$, respectively), while the 50 mg/kg dose showed a substantial but non-significant improvement. Overall, these findings indicate that ECT possesses anxiolytic activity, with higher doses demonstrating greater efficacy, although fluoxetine remained the most effective treatment (Seibenhener & Wooten, 2015).

The findings confirm that anxiety-like behaviour was successfully induced, as demonstrated by reduced centre exploration in the OFT, while untreated rats showed no spontaneous behavioural recovery. Fluoxetine produced the most pronounced anxiolytic effect, significantly increasing centre exploration, consistent with its well-established mechanism of enhancing serotonergic neurotransmission. Similarly, treatment with the extract of clove and turmeric (ECT) improved exploratory behaviour, with the 100 and 200 mg/kg doses producing statistically significant anxiolytic-like effects, whereas the 50 mg/kg dose, despite showing a large percentage increase, did not achieve statistical significance due to greater variability among animals. The observed anxiolytic effects of ECT are likely attributable to the synergistic actions of its major phytochemicals, eugenol and curcumin, which possess antioxidant, anti-inflammatory, neuroprotective, and

monoaminergic modulatory properties. These mechanisms may reduce oxidative stress and neuroinflammation associated with anxiety, thereby improving exploratory behaviour. Overall, the results indicate that ECT exhibits dose-dependent anxiolytic-like activity, with higher doses demonstrating greater efficacy and effects approaching those of fluoxetine (Ceremuga *et al.*, 2017).

The Tail Suspension Test (TST) is widely recognized for its reliability in evaluating antidepressant activity. It is a widely accepted behavioral model for assessing antidepressant-like activity in rodents. It is based on the principle that rodents, when suspended by their tails, initially exhibit escape-oriented behaviors, which gradually transition into immobility. A reduction in immobility time following treatment is considered indicative of an antidepressant-like effect (Can *et al.*, 2012).

From (Table 2) of the present study, the immobility time which serve as a key indicator of behavioral despair, was assessed at three different phases: the baseline (before induction of depression), the post-induction (after induction of the depression), and the post-treatment (after administration of extracts of interest). The results showed that depressive-like behaviour was successfully induced, as reflected by the marked increase in immobility time in all treatment groups following induction. Fluoxetine produced the greatest antidepressant-like effect, significantly reducing immobility by 86.9% ($p < 0.01$). Treatment with the extract of clove and turmeric (ECT) also reduced immobility in a dose-dependent manner. Although the 50 and 100 mg/kg doses produced substantial reductions (71.6% and 70.2%, respectively), these effects were not statistically significant. In contrast, the 200 mg/kg dose significantly reduced immobility by 85.1% ($p < 0.01$), achieving an effect comparable to fluoxetine.

The present study confirmed the immobility of the combined clove and turmeric extracts as reported by several studies (Hassan and Rahman, 2024). Fluoxetine (a selective serotonin reuptake inhibitor, SSRI) was used as the positive control. Fluoxetine exerts its antidepressant effects by increasing serotonin levels in the brain, which enhances mood and motivation (Stenfors *et al.*, 2001).

The results showed that treatment with the extract of clove and turmeric (ECT) reduced immobility time in a dose-dependent manner. Although the 50 and 100 mg/kg doses produced substantial reductions (71.6% and 70.2%, respectively), only the 200 mg/kg dose achieved a statistically significant effect. Notably, the antidepressant-like effect of ECT 200 mg/kg (85.1%

reduction) was comparable to that of fluoxetine, with both groups showing similarly low post-treatment immobility times. The antidepressant-like activity of ECT may be attributed to the combined effects of eugenol from clove and curcumin from turmeric, which are known for their antioxidant, anti-inflammatory, and neuroprotective properties. These phytochemicals may help reduce oxidative stress and neuroinflammation while improving neuronal function, thereby contributing to the observed behavioural improvements. Overall, the findings suggest that ECT exhibits dose-dependent antidepressant-like activity, with the 200 mg/kg dose providing the most consistent and effective response, approaching the efficacy of fluoxetine.

The Forced Swim Test (FST) is one of the most commonly used behavioral assays to evaluate antidepressant-like activity in rodents. In this test, animals are placed in an inescapable container filled with water, and their immobility time is recorded. Reduced immobility time after treatment suggests an increase in motivation and a decrease in behavioral despair, which is a hallmark of antidepressant activity (Can *et al.*, 2012).

From (Table 3), provided additional evidence for the test substance's antidepressant-like effects. The Forced Swim Test findings indicate that the experimental groups generally had comparable baseline immobility times, while the marked increase in immobility after induction in the FLX- and ECT-treated groups suggests the development of depressive-like behaviour. Following treatment, distilled water produced no meaningful improvement, whereas fluoxetine significantly reduced immobility by 69.9% ($p < 0.01$), confirming its antidepressant-like activity. ECT also reduced immobility, with reductions of 50.2%, 58.2%, and 65.9% at 50, 100, and 200 mg/kg, respectively. However, only the 100 mg/kg ($p < 0.05$) and 200 mg/kg ($p < 0.01$) doses produced statistically significant effects, while the 50 mg/kg dose was not significant. Numerous studies have confirmed the neuroprotective potentials of both clove and turmeric in management of neurodegenerative pathophysiology like oxidative stress, neuroinflammation, mitochondrial dysfunction, tau protein abnormal aggregation, neurofibrillary tangle, acetylcholine deficiency, and hormonal imbalance (Yu *et al.*, 2002; Sanmkhani *et al.*, 2022).

The antidepressant-like effects observed in this study may be attributed to the presence of curcumin in turmeric and eugenol in clove, both of which have been shown to modulate monoamine

neurotransmitter systems (Xu *et al.*, 2005; Amaral *et al.*, 2013). The dose-dependent nature of the response aligns with previous studies indicating that higher doses of curcumin and eugenol result in greater antidepressant-like effects in animal models (Xu *et al.*, 2005; Hewlings & Kalman, 2017). However, the potential biphasic effects of plant-derived compounds should also be considered, as excessively high doses may alter neurotransmitter balance or induce sedative effects that counteract antidepressant benefits (Amaral *et al.*, 2013).

Clove (*Syzygium aromaticum*) mitigates depression-like behaviors associated with diazepam by acting as a strong antioxidant, reducing oxidative stress, and acting on neurochemical pathways such as GABA and serotonin, effectively reversing deficits in memory and mood (Parvizi *et al.*, 2022). Clove extracts can increase the production of serotonin and influence acetylcholine and dopamine pathways, helping to counteract the depressive effects of benzodiazepines. The Clove oil reduces oxidative stress in the brain, which is a mechanism often involved in depression (Garabadu & Sharma 2019). The active compound of turmeric (Curcumin) has been found to prevent cognitive impairment by protecting hippocampal and frontal lobe structure and modulating the brain's inflammatory response to drug-induced stress and depression (Sevastre Berghian *et al.*, 2016). The treatment of diazepam induced depression like effect in the albino rats (Haider *et al.*, 2019) by the combined extracts of clove and turmeric, demonstrated reduction in immobility time significantly in both FST and TST (Sheethal *et al.*, 2020).

These findings suggest a dose-dependent effect of the extracts, where lower doses may exert anxiolytic properties, while higher doses could lead to diminished efficacy or even adverse effects. This phenomenon is not uncommon in phytochemical research, as certain compounds may exhibit biphasic effects depending on the concentration administered. For instance, studies have reported that curcumin, the active component in turmeric, can have varying biological effects at different doses (Hewlings and Kalman, 2017).

These findings further support the synergistic effects of medicinal plants used by various societies globally either as spices in preparation of daily foods or as medicinal purposes (Verma, 2016). Turmeric contains curcumin, which has been shown to possess antidepressant properties, potentially through the modulation of neurotransmitter levels and anti-inflammatory effects (Lopresti *et al.*, 2012). Clove is

rich in eugenol, a compound with known anxiolytic and antidepressant activities, possibly due to its interaction with the central nervous system and antioxidant properties (Cortés-Rojas *et al.*, 2014).

CONCLUSION

This study provides compelling evidence of the anxiolytic and antidepressant-like properties of the test substance, particularly at higher doses. The results from the OFT, TST, and FST collectively support its potential as a novel therapeutic agent for managing anxiety and depression. While further research is needed to fully elucidate its mechanisms and optimize its clinical application, the findings represent an important step toward the development of new treatments for these debilitating conditions. Despite the promising results, the study is not without limitations. The small sample size (n=5 per group) may reduce the statistical power of the findings and limit their generalizability. Moreover, the absence of molecular or biochemical analyses restricts the ability to draw conclusions about the underlying mechanisms of action. Future studies should address these limitations by including larger sample sizes and incorporating advanced techniques, such as high-performance liquid chromatography (HPLC) for neurotransmitter quantification or immunohistochemistry for receptor expression analysis.

AUTHOR CONTRIBUTION: YHAG and FSD developed and designed the study. GAY and YHAG performed the experiments, UBK analyzed the data. YHAG and FSD drafted the manuscript. All authors reviewed and approved the final version of the manuscript.

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