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Evaluation the Aqueous Leaf Extract and Solvent Fractions of *Pyrenacantha staudtii* Engl. (Icancinaceae) for Antidepressant and Anxiolytic-like Activities in Mice

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ABSTRACT

Background and Purpose: *Pyrenacantha staudtii* Engl. (Icacinaceae) is a plant used traditionally for treating blennorrhea, hernia, insomnia, intestinal pain and diarrhea in Nigeria. The aim of the present study was to screen its aqueous extract and fractions for some neuropharmacological activities.

Materials and Methods: Classical behavioral tests for antidepressant-like and anxiolytic-like, and hypnotic-sedative activities were conducted using the crude aqueous extract of *Pyrenacantha staudtii* (CAEPS) and its solvent fractions at doses of 100, 200, and 400 mg/kg, intraperitoneally.

Results: CAEPS at all doses significantly reduced the immobility time of mice in force swim test (FST) but reduced only at 400 mg/kg dose in tail suspension test (TST). The dichloromethane fraction (DCMF) and n-hexane (n-HF) did not significantly alter the results in both models. The ethanol fraction (EF) at 400 mg/kg and the ethyl acetate fraction (EAF) significantly reduced immobility time in FST and TST. CAEPS and EF at all doses, and EAF at 400 mg/kg significantly reduced the number of lines and squares crossed in open field test, while DCMF and n-HF did not. The number of head dips significantly decreased in the extract-treated groups in the hole board test. The CAEPS at 400 mg/kg and EF at 200 and 400 mg/kg doses significantly increased the time spent in the open arms of the elevated plus maze (EPM) but EAF, DCMF, and n-HF did not. CAEPS and EF at all doses and EAF at 400 mg/kg prolonged phenobarbitone-induced sleeping time while DCMF did not.

Conclusion: The extract and polar fractions possess antidepressant-like, anxiolytic-like and sedative properties. This lends credence to the use of the plant in the treatment of mood disorders in Nigerian traditional medicine.

INTRODUCTION

The use of drugs for the relief of stress, tension, and anxiety is well established. Benzodiazepines and other drugs that act on neurotransmitter systems are currently being used (Zhang and Yao, 2019). However, ineffective treatment and undesirable effects such as dependence still exist in many cases.

Pyrenacantha staudtii (Engl.) Engl. (Icacinaceae) is called *Ahara* in Yoruba, *Nhia* in Igbo and *Ohogbo* in Edo, languages of Nigeria. It is widely distributed in tropical Africa such as secondary jungles of Southern Nigeria, Western Cameroon, and across Central Africa (Daziel, 1948). Traditionally, the plant is used for the treatment of blennorrhea, intestinal pain and hernia (Daziel, 1948; Oliver-Bever, 1986; Iwu, 1993), and diarrhea. The leaf extract has been reported for its anti-inflammatory and antiulcerogenic activities (Akubue *et al*., 1983), smooth muscle relaxant activity (Aguwa and Okunji, 1986), anticonvulsant, and analgesic activities (Awe *et al*., 2005). The plant extract was reported to contain saponins and alkaloids (Okunji and Iwu, 1988). In the present study, we screened the plant for some neuropharmacological effects using murine models.

MATERIALS AND METHODS

Plant Material

Fresh leaves of *Pyrenacantha staudtii* Engl. (Icacinaceae) were collected at Iwo Local Government Area, Osun State, in the southern part of Nigeria, in June 2024. The leaves were identified by Mr. T.K. Odewo, a taxonomist at the herbarium of the Federal Institute of Forest Research Ibadan, where a voucher specimen with number 106886 was deposited.

Preparation of Plant Extract

The fresh leaves (1 kg) were air-dried at room temperature. The air-dried leaves were milled to fine powder using an electric blender. The powdered leaf was macerated twice in 2.5 L of distilled water at room temperature for 48 h (with occasional shaking). The combined aqueous extracts were filtered and concentrated to dryness under reduced pressure at 60±1°C in a rotary evaporator. Freeze-drying and solvent elimination of the resulting aqueous extract yielded 60 g (i.e., 6.0 % w/w yields) of a light brown, powdery crude aqueous extract of Pyrenacantha staudtii leaf (CAEPS). Without further purification, aliquots of the crude extract were weighed and dissolved in distilled water (at room temperature) for use on each day of experiments.

Fractionation of CAEPS

CAEPS was suspended in water and subsequently extracted with solvents of increasing polarity, namely n-hexane (250 mL), dichloromethane (250 mL), ethyl acetate (250 mL) and ethanol (250 mL). Each fraction was concentrated under reduced pressure to yield n-hexane fraction (n-HF), dichloromethane fraction (DCMF), ethyl acetate fraction (EAF), and ethanol fraction (EF).

Animals

Healthy male and female mice weighing 20 – 25 g were used for this study. The animals were kept and maintained under natural laboratory conditions of temperature, humidity, and light. They were allowed free access to food (standard pellet) and tap water. The animals were assigned to experimental groups consisting of aqueous extract and fractions, reference drug-treated, and distilled water-treated. All the animals were fasted for 16 h but allowed free access to tap water before the commencement of the experiments.

Neuropharmacological Tests

Forced swim test

The animals were assigned to five groups (n=5). Group 1 animals were treated with normal saline (10 mL/kg), groups 2, 3, and 4 received the CAEPS at doses of 100, 200, and 400 mg/kg, i.p.) respectively, while group 5 received imipramine (20 mg/kg, i.p.) as the standard drug. A glass cylinder (26 cm high, 12 cm in diameter) was filled to a depth of 16 cm with water and each mouse was placed in the cylinder for 6 min. The duration of immobility during the last 5 min was recorded (Porsolt *et al*., 1977; Ren *et al*., 2016). The same procedure was repeated for the fractions (EF, EAF, DCMF, and n-HF) at doses of 100, 200, and 400 mg/kg.

Tail suspension test

The mice were assigned to five groups (n=5). Group 1 animals were treated with normal saline (10 mL/kg), groups 2, 3, and 4 received the CAEPS at doses of 100, 200, and 400 mg/kg, i.p., respectively, while group 5 received imipramine (20 mg/kg, i.p.). All the mice were subjected to tail suspension test 30 min after the treatment. Each mouse was suspended on the border of a table using a tape with an adhesive that was positioned about 1.5 cm from the tip of its tail. The tail suspension was for 6 min but motionlessness (duration of immobility) was only scored during the last 5 min (Salako *et al*., 2019). The same procedure was repeated for the fractions (EF, EAF, DCMF, and n-HF) at doses of 100, 200, and 400 mg/kg.

Open field test

The animals were grouped as in FST except that diazepam was used as the standard. The center of the wooden box used for the test was separated into 8 × 8 cm with 16 squares and the dimension was 50 × 50 × 25 cm. Each mouse was placed on the surrounding zone 30 min after each group was intraperitoneally treated with CAEPS at doses of 100, 200, and 400 mg/kg, distilled water

(10 mL/kg), or diazepam (1 mg/kg). The number of lines and squares crossed including rearing and grooming were counted and recorded for 5 min (Zhou *et al*., 2019). The same procedure was repeated for the fractions (EF, EAF, DCMF, and n-HF) at doses of 100, 200, and 400 mg/kg.

Hole-board test

The animals were grouped as in FST above, but diazepam was used as the standard. The board (42 × 42 × 30 cm) consists of 16 equidistant holes (2.5 cm in diameter). The floor of the box was positioned 15 cm above the ground. Thirty minutes after the treatment with normal saline (10 mL/kg), CAEPS at doses of 100, 200, and 400 mg/kg, i.p., and diazepam (1 mg/kg, i.p.), the exploratory behavior of the mice (number of head-dips and squares crossed with all four paws) were counted and recorded for 5 min (Takeda *et al*., 1998). The same procedure was repeated for the fractions (EF, EAF, DCMF, and n-HF} at doses of 100, 200, and 400 mg/kg.

Elevated plus maze test

The experiment was conducted with the maze placed 50 cm above ground. Mice (5 per group) were grouped as stated for FST. They were given normal saline (10 mL/kg, control) orally. CAEPS was administered intraperitoneally at doses of 100, 200, and 400 mg/kg and diazepam (1 mg/kg) 30 min before the test. Each mouse was positioned on the platform and observed for 5 min. For each mouse, the parameters recorded were number of entries and time spent in the open arms (Lister, 1987). The same procedure was repeated for the fractions (EF, EAF, DCMF, and n-HF) at doses of 100, 200, and 400 mg/kg.

Phenobarbitone-induced sleep test

The method used by Bourin (1986) was adopted. Briefly, the animals were assigned to 5 groups of 5 mice each. Group 1 animals were treated with normal saline (10 mL/kg), while groups 2, 3, and 4 received the CAEPS at doses of 100, 200, and 400 mg/kg, i.p., respectively. The standard drug diazepam (1 mg/kg, i.p.) was used as positive control group. All pretreatments were done intraperitoneally 30 min before administration of phenobarbitone sodium at a dose of 40 mg/kg. Time interval between loss and regain of righting reflex was taken as index of hypnosis. Duration of sleep was considered completed when mice did not accept the decubitodorsal position on three consecutive trials (Goyal and Sasmal, 2014; Fageyinbo *et al*., 2019). The same procedure was repeated for the fractions (EF, EAF, DCMF, and n-HF) at doses of 100, 200, and 400 mg/kg.

Statistical Analysis

Statistical analysis was performed using one-way analysis of variance (ANOVA), followed by Dunnett's multiple comparison test. Data obtained from the study are expressed as mean ± SEM (standard error of mean). P-values less than 0.05 were considered significant.

RESULTS

Yield by the Solvent Fractions

The yield by the fractions obtained from crude aqueous extract of *Pyrenacantha staudtii* were n-HF: 5.7% w/w; DCMF: 13.8 w/w; EAF: 20 % w/w; and EF: 26.2 % w/w).

Effect of CAEPS and Fractions on Mice Behavior in Tail Suspension and Forced Swim Tests

In the tail suspension test, the extract at 400 mg/kg significantly reduced the duration of immobility by mice (51.24±2.67 s) compared to the control (145.17±3.23 s) and the standard drug, imipramine (81.43±2.45 s). In the forced swim test, the duration of immobility by the mice decreased in a dose-dependent manner. EF and DCMF had similar effects with the extract at a 400 mg/kg dose in both tests. However, DMF and n-HF did not significantly affect the results of both models as shown in Table 1.

Table 1: Effect of CAEPS and fractions on tail suspension and forced swim tests.

Treatments	Doses (mg/kg)	Immobility Time (s)**	
		TST	FST
Control	0	145.17±3.23	139.24±2.75
CAEPS	100	133.29±4.15	95.23±3.34*
	200	132.53±2.64	90.23±2.25*
	400	51.24±2.67*	79.41±1.17*
EF	100	143.21±3.05	140.43±3.12
	200	140.03±2.76	138.07±4.06
	400	67.14±1.56*	51.03±2.35*
EAF	100	145.01±5.01	137.31±3.51
	200	144.32±6.16	138.11±4.06
	400	71.63±1.21*	76.42±3.41*
DCMF	100	144.13±4.41	136.04±3.21
	200	145.65±7.02	137.13±4.01
	400	141.23±3.07	138.21±3.96
n-HF	100	145.31±6.34	136.34±3.46
	200	145.07±5.51	136.01±3.07
	400	143.91±4.09	135.71±3.42
Imipramine	20	81.43±2.45*	75.31±2.45*

**Values are mean ± SEM (n=5).

*Values are statistically significant (P<0.05) compared with control using one-way ANOVA followed by Dunnett's post-hoc test.

Effect of CAEPS and Fractions on the Behavior of Mice in an Open Field

In the open field test, the CAEPS at doses of 100, 200, and 400 mg/kg significantly caused a reduction in the number of lines and squares crossed, rearing and grooming behavior compared with the control (Table 2). Similarly, EF at the doses of 100, 200, and 400 mg/kg reduced the number of lines and squares crossed, rearing and grooming behavior. EAF also caused a similar effect but at a dose of 400 mg/kg.

DCMF and n-HF did not significantly influence the behavior of mice on the open field. Compared with control, diazepam reduced the number of rearing, grooming, lines, and squares crossed.

Table 2: Effect of CAEPS and fractions on the activity of mice in the open field tests.

Treatments	Doses (mg/kg)	Rearing	Number of Line Crossing**	Number of Square Crossing**	Grooming**
Control	0	5.12±0.87	95.31±2.24	5.45±0.68	4.15±0.75
CAEPS	100	3.61±0.52	63.54±3.21*	3.86±0.34*	2.31±0.81*
	200	2.01±0.18*	57.32±5.32*	1.78±0.41*	1.24±0.26*
	400	0.65±0.30*	39.27±3.23*	0.93±0.34*	0.61±0.18*
EF	100	4.02±1.01*	87.61±4.02*	4.61±1.05*	4.01±1.52
	200	2.75±0.81*	61.31±2.13*	3.11±0.95*	2.81±0.59*
	400	0.91±0.57*	41.21±3.02*	1.07±0.71*	1.01±0.32
EAF	100	5.21±1.02	94.23±4.05	5.01±1.58	4.21±1.52
	200	5.01±0.31	93.71±3.45	4.71±0.65	4.13±0.89
	400	2.83±0.45*	43.22±2.32*	1.81±0.76*	1.22±0.91*
DCMF	100	5.11±1.34	94.71±4.50	5.41±1.35	4.61±1.31
	200	4.90±1.12	94.53±4.02	5.43±1.02	4.12±1.53
	400	5.06±1.32	93.71±3.74	4.91±1.31	4.17±1.21
n-HF	100	5.00±1.02	94.35±4.45	5.22±1.50	4.23±1.42
	200	4.87±1.21	94.41±4.04	5.01±1.12	4.01±1.02
	400	4.95±0.87	95.01±3.32	5.13±1.67	3.97±0.97
Diazepam	1	1.31±0.71	32.28±2.45	1.17±0.41*	2.03±0.16*

**Values are mean ± SEM (n=5).
*Values are statistically significant (P<0.05) compared with control using one-way ANOVA followed by Dunnett's post-hoc tests.

Effect of CAEPS and Fractions on the Hole Board

Mice that received CAEPS at doses of 100, 200, and 400 mg/kg had significantly decreased head dip counts compared to control. Similarly, those that had EF at 200 and 400 mg/kg doses had decreased head dip counts compared with control. The dose of 200 mg/kg EAF similarly decreased the head dip counts. DCMF and n-HF did not significantly decrease the head dip counts. CAEPS and EF at 400 mg/kg dose showed a comparable effect with diazepam.

Table 3: Effect of CAEPS and its fractions on head dips count by mice on the hole board

Treatments	Doses (mg/kg)	Head-dip Counts**
Control	0	16.41 ± 2.43
CAEPS	100	7.51 ± 1.23*
	200	5.23 ± 1.05*
	400	3.45 ± 0.76*
EF	100	15.49 ± 1.73
	200	9.42 ± 1.05*
	400	4.12 ± 1.35*
EAF	100	16.05 ± 2.40
	200	7.97 ± 1.67*
	400	15.31 ± 2.03
DCMF	100	16.21 ± 2.13
	200	16.01 ± 1.79
	400	15.23 ± 1.47
n-HF	100	15.01 ± 2.20
	200	16.11 ± 2.49
	400	15.67 ± 2.12
Diazepam	1	3.89 ± 0.53*

**Valu es are mean ± SEM (n=5).
*Values are statistically significant (P<0.05) in relation to control using one-way ANOVA followed by Dunnett's post-hoc tests

Table 4: Effect of CAEPS and its fractions on phenobarbitone-induced sleep test in mice.

Treatments	Doses (mg/kg)	Onset of Sleep (min)**	Duration of Sleep (min)**
Control	0	14.42±1.23	211.43±3.46
CAEPS	100	13.41±0.23	192.35±4.57
	200	10.32±1.07*	240.52±3.64*
	400	6.05±0.21*	301.24±5.31*
EF	100	11.12±2.01*	201.45±4.21
	200	10.13±1.81*	258.13±5.45*
	400	8.25±1.01*	289.61±4.92*
EAF	100	14.15±2.31	207.42±3.42
	200	10.11±1.21*	296.14±4.47*
	400	9.01±1.25*	323.31±5.68*
DCMF	100	14.27±2.11	201.95±3.59
	200	14.29±1.59	221.09±4.01
	400	14.12±2.03	210.35±5.72
n-HF	100	14.16±2.15	211.40±4.01
	200	13.49±1.95	208.11±4.74
	400	13.92±2.01	217.35±4.01
Diazepam	1	8.23±0.67*	338.43±4.27*

**Values are mean ± SEM (n=5).
*Values are statistically significant (P<0.05) in relation to control using one-way ANOVA followed by Dunnett's post-hoc tests

Effect on CAEPS and Fractions on Phenobarbitone-induced Sleep

CAEPS at doses of 100, 200, and 400 mg/kg, and EF and EAF at doses of 200 and 400 mg/kg significantly (P<0.05) shortened the onset but prolonged the duration of phenobarbitone-induced sleep, in mice. Doses of DCMF and n-HF did not significantly reduce the onset nor prolong phenobarbitone-induced sleeping time in mice (Table 4)

Effect of CAEPS and Fractions on the Performance of Mice on the Elevated Plus Maze

CAEPS at doses of 200 and 400 mg/kg, EF and EAF at a dose of 400 mg/kg significantly increased the time spent on the open arms compared with the control. DCMF and n-HF did not significantly influence the movement of mice in the elevated plus maze as shown in Table 5.

Table 5: Effect of CAEPS and Fractions on the behavior of mice on the elevated plus maze.

Treatments	Doses (mg/kg)	Time Spent (s)		Number of Entries	
		Open Arms**	Closed Arms**	Open Arms**	Closed Arms**
Control	0	130.75±7.15	191.27±4.05	5.02±0.05	20.21±1.02
CAEPS	100	131.34±4.41	160.03±5.21	11.13±1.01	17.14±1.03
	200	195.21±4.23*	145.21±3.25	12.35±1.62	9.34±0.09
	400	210.41±6.54*	129.31±4.32	11.25±1.23	7.48±0.16
EF	100	131.23±3.21	189.45±5.45	10.71±1.02	19.14±1.29
	200	130.56±4.32	190.35±3.43	12.78±1.21	17.37±1.21
	400	212.38±5.24*	135.62±4.31*	11.45±1.32	9.94±1.35*
EAF	100	129.36±3.41	190.53±4.02	12.54±1.63	17.34±2.09
	200	129.87±3.65	187.37±4.54	11.89±1.43	18.41±2.12
	400	207.52±5.67*	145.91±3.62*	11.48±1.35	9.72±0.12*
DCMF	100	130.01±3.71	190.38±4.71	12.91±1.68	19.64±2.11
	200	129.67±3.37	188.74±4.37	11.78±1.58	19.75±1.23
	400	130.46±3.52	191.45±4.65	12.05±1.51	20.24±2.35
n-HF	100	129.68±3.49	193.01±5.02	12.81±1.97	19.86±2.75
	200	130.75±3.23	189.51±4.32	12.21±1.09	20.18±3.03
	400	129.01±3.61	190.61±3.58	11.95±1.14	19.57±2.43
Diazepam	1	201.35±5.47*	98.71±2.23	24.21±2.03*	8.47±0.10*

**Values are mean ± SEM (n=5).

*Values are statistically significant (P<0.05) in relation to control using one-way ANOVA followed by Dunnett's post-hoc tests

DISCUSSION

The crude aqueous extract of *Pyrenacantha staudtii* (CAEPS) and fractions were evaluated for CNS activities in mice using tail suspension and forced swim tests (antidepressant-like effect), open field, hole board, elevated plus maze tests (anxiolytic-like effect) and pentobarbitone sodium-induced sleep time test (sedative effect). The extract and its fractions worked like mirtazapine an antidepressant that also has sedative and anxiolytic effects, meaning it can help reduce anxiety and promote relaxation (Mohammad-Reza Zarrindast *et al*., 2024). Forced swim and tail suspension tests place mice in an inescapable stressful situation, an action that produces a mental condition of hopelessness and despair that could be accounted for in the form of immobility (Yan *et al*., 2015; Dhamija *et al*., 2017). Immobility or despair behavior produced in both forced swim and tail suspension tests are taken as a prototype of depression which reflects the behavioral despair seen in depressed people (Afsar *et al*., 2017). In general, the immobility response is reduced by the administration of antidepressant drugs and other treatments that are therapeutically effective in depression (Lucki, 2010). In this study, the extract at a dose of 400 mg/kg significantly reduced the duration of immobility by mice, while in the forced swim test, the duration of immobility by mice decreased dose-dependently. However, the dichloromethane and n-hexane fractions did not significantly affect either model, while ethanol and ethyl acetate fractions at the dose of 400 mg/kg considerably reduced the duration of immobility in FST and TST. The extract and fractions reduced the duration of immobility in the two tests, suggesting an antidepressant-like effect.

In the open field test, the number of line crosses and the frequency of rearing are used to measure locomotor activity, exploration, and anxiety (Zimcikova *et al*., 2017). In addition, locomotor activity is considered to measure the level of excitability of the CNS (Nahar *et al*., 2012). The study revealed that CAEPS significantly and dose-dependently reduced the number of lines, squares crossed, rearing, and grooming behavior compared with control. Similarly, all the doses of the ethanol fraction reduced the

number of lines, number of squares crossed, rearing, and grooming behavior dose-dependently. The ethyl acetate fraction also has a similar effect but at a dose of 400 mg/kg. The dichloromethane and n-hexane fractions did not significantly affect the behavior on the open field. The standard drug **(**diazepam) gave results similar to the extract and fractions but was more effective than either the extract or fractions. The reduction in these parameters suggests that the extract and fractions could depress the CNS, which could be related to a sedative effect as previously reported (Nahar *et al* 2012; Cryan *et al*., 2005; Yan *et al*., 2015; Sobreira Dantas Nóbrega de Figuêiredo *et al*. 2019).

The effect of the extract was also evaluated in the hole-board test. The test assesses anxiety, emotionality, and stress response (Takeda *et al*., 1998; Wei *et al*., 2007). A significant attenuation in head dips during the exploratory behavior is a measure of anxiolytic-like effect (File and Wardill, 1975). In this model, the extract and ethanol fraction significantly decreased the number of head dips by mice compared to the control. This observation suggests the depressant effect of the extract.

In the elevated plus maze, anxiety is indicated by an animal's avoidance of the open arms when placed in the maze. An anxiolytic-like effect is indicated by an increase in the time spent by the animals in the open arm. The anxiolytic-like effect of the extract and fractions showed in the form of longer (cumulative) time spent in the open arm of the maze (Rang *et al*., 2003; Foyet *et al*., 2012).

The central nervous system depressant activity of the plant extract and ethanol fraction was also evident by their ability to enhance phenobarbitone-induced hypnosis in a dose-dependent manner. The extract and ethanol fraction were able to significantly shorten the onset and prolong the duration of phenobarbitone-induced sleep in mice thus confirming the sedative properties of the plant (Emamghoreishi and Heidari-Hamedani, 2006).

Endogenous neurotransmitters in the brain, particularly acetylcholine, dopamine, norepinephrine, serotonin, GABA, histamine, and neuropeptides, have been shown to play significant roles in sleep mechanisms (Prakasha *et al*., 2008; Howlader *et al*., 2017). The effects of the extract on phenobarbitone-induced sleep in mice may be attributed to its potential interactions with some of these central neurotransmitters that are involved in sleep regulation (Howlader *et al*., 2017).

Phytochemical studies on the plant have revealed the presence of some phytoconstituents, such as flavonoids, terpenoids, tannins, and steroids. Flavonoids, like apigenin, hesperidin, and certain citrus flavonoids, are reported to have behavioral effects in different animal models of anxiety and sedation. The CNS depressant action of some flavonoid glycosides such as hesperidin, naringin, and rutin in mice was reported by Fernandez *et al*. (2006). The sedative and anxiolytic actions of terpenoids have been reviewed (Parmar *et al*., 2013). Therefore, the CNS depressant activity of the extract and fraction may be due to the presence of these constituents.

CONCLUSION

In conclusion, the aqueous extract of *Pyrenacantha staudtii* leaf and ethanol fraction possess anxiolytic-like, antidepressant-like and sleep enhancing properties. This finding suggests that the polar phytoconstituents were responsible for the observed effect. Our results support the use of the plant in traditional medicine.

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CONFLICT OF INTEREST

The authors declare that they have no known competing financial interests or personal relationships that could have influenced the work reported in this article.

AUTHORS' CONTRIBUTION

OKW and DOK designed the study; MKB and AFA analyzed the data; and OKW and DOK drafted the manuscript. All authors read and approved the manuscript for publication.

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AUTHORS' DECLARATION

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

REFERENCES

Afsar T, Razak S, Khan MR, Almajwal A (2017). Anti-depressant and anxiolytic potential of *Acacia hydaspica* R. Parker aerial parts extract: Modulation of brain antioxidant enzyme status *BMC Complementary Alternative Medicine* 17: 228

Aguwa CN, Okunji CO (1986). Gastrointestinal studies of *Pyrenacanthia staudtii* leaf extract. *Journal of Ethnopharmacology* 5: 45-55.

Akubue PI, Mittal GC, Aguwa CN (1983). Preliminary pharmacological study of some Nigerian medicinal plants. *Journal of Ethnopharmacology* 8(1): 53-63.

Awe EO, Makinde JM, Olajide OA, Wakeel OK, Adeloye AO (2005). Evaluation of analgesic, anticonvulsant and hypnotic activities of *Pyrenacanthia staudtii*. *African Journal of Traditional, Complementary and Alternative Medicine* 2: 122-128.

Bouri M, Poisson L, Larousse C (1986). Piracetam interaction with neuroleptics in psychopharmacological tests. *Neuropharmacology* 19: 93-96.

Cryan JF, Mombereau C, Vassout A (2005). The tail suspension test as a model for assessing antidepressant activity: Review of pharmacological and genetic studies in mice *Neuroscience Biobehavioural Review* 29: 571-625.

Daziel JM (1948). The useful plants of West Tropical Africa. London Crown Agents for Overseas Government and Administration. London. Mill Bank, p. 45.

Dhamija I, Parle M, Kumar S (2017). Antidepressant and anxiolytic effects of *Garcinia indica* fruit rind via monoaminergic pathway. 3 Biotech 7(2): 131. doi: 10.1007/s13205-017-0766-x.

Emamghoreishi M, Heidari-Hamedani G (2006). Sedative-hypnotic activity of extracts and essential oil of coriander seeds. *Iran Journal of. Medicinal Sciences* 31(1): 22-27.

Fageyinbo MS, Odumeso AO, Rotimi K, Fasipe OJ, Olaye mi SO (2019). Anticonvulsant, anxiolytic and sedative activities of hydro-ethanolic leaf extract of *Mitragyna stipulosa*. *Journal of Herbs Spices Medicinal Plants* 25: 43-54.

File SE, Wardill AG (1975). Validity of head-dipping as a measure of exploration in a modified hole-board *Psychopharmacologia* 44 (1): 53-59.

Fernandez SP, Wasowski C, Loscalzo LM, Granger RE, Johnston GAR, Paladini AC, Marder M, (2006). Central nervous system depressant action of flavonoid glycosides. *European. Journal of Pharmacology* 539: 168-176.

Foyet HS, Tsala DE, Bouba AA, Hritcu L (2012). Anxiolytic and antidepressant-like effects of the aqueous extract of *Alafia multiflora* stem barks in rodents. Advances in Pharmacological Sciences 2012: 912041. doi: 10.1155/2012/912041.

Goyal M, Sasmal D (2014). CNS depressant and anticonvulsant activities of the alcoholic extract of leaves of *Ziziyphus nummularia Journal of Ethnopharmacology* 151(1): 536-542.

Howlader SI, Siraj A, Dey SK, Hira A, Ahmed, A, Hossain H (2017). *Ficus hispida* bark extract prevents nociception, inflammation, and CNS stimulation in experimental animal model. Evidence *Based Complementary Alternative Medicine* 2017:7390359. doi: 10.1155/2017/7390359.

Iwu MM (1993). Handbook of African Medicinal Plants. New York. CRC Press Inc, p. 27.

Lister RG (1987). The use of a plus-maze to measure anxiety in the mouse *Psychopharmacology* 92: 180-185.

Lucki I (2010). Behavioral Despair. Stolerman IP (Ed.), Encyclopedia of Psychopharmacology, Springer, Berlin, Heidelberg. 202-204

Zarrindast MR, Hajikarimloo B, Raissi-Dehkordi N, Raissi-Dehkordi N, Khakpai F (2024). Preclinical evidence for the anxiolytic- and antidepressant-like effects of citicoline and imipramine in the sciatic nerve-ligated mice. IBRO Neuroscience Reports 17: 364-371. doi: 10.1016/j.ibneur.2024.10.007.

Nahar L, Zahan R, Morshed MTI, Haque A, Alam Z, Mos addik A (2012). Antioxidant, analgesic and CNS depressant effects of Synedrella nodiflora *Pharmacognosy Journal*, 4 (31): 29-36

Okunji CO, Iwu MM (1986). Control of schistosomiasis using Nigerian medicinal plants. *Annual Review of Nutrition* 6: 433-458.

Oliver-Bever B (1986). Medicinal Plants in Tropical West Africa. London. Cambridge University Press, p. 61.

Parmar SK, Sharma TP, Airao, VB, Bhatt R, Aghara R, Chavda S, Rabadiya, SO, Gangwal AP (2013). Neuropharmacological effects of triterpenoids. *Phytopharmacology* 4(2): 354-372.

Porsolt RD, Bertin A, Jalfre M (1977). Behavioral despair in mice: A primary screening test for antidepressants. *Archives Internationales de Pharmacodynamie et de Thérapie* 229(2): 327-336.

Prakash T, Rao NR, Swamy AH (2008). Neuropharmacological studies on *Wedelia calendulacea* Less stem extract. Phytomedicine 15(11): 959-970. doi: 10.1016/j.phymed.2008.05.005.

Rang HP, Dale JM, Ritter MM (2003). Pharmacology (5th ed.), Churchill Livingstone.

Ren L, Tao W, Zhang H, Xue W, Tang J, Wu R, Xia B, Wu H, Chen G (2016). Two standardized fractions of *Gardenia jasminoides* Ellis with rapid antidepressant effects are differentially associated with BDNF up-regulation in the hippocampus. Journal of Ethnopharmacology 187: 66-73. doi: 10.1016/j.jep.2016.04.023.

Salako OA, Akindele AO, Balogun OO, Adeyemi OO (2019). Investigation of antidepressant, anxiolytic and sedative activities of the aqueous leaf extract of *Musa sapientum* Linn. (Banana; Musaceae) *Drug Research (Stuttg)* 69(3): 136-143.

Sobreira Dantas Nóbrega de Figuêiredo FR, Monteiro ÁB, Alencar de Menezes IR, Sales VDS, Petícia do Nascimento E, Kelly de Souza Rodrigues C, Bitu Primo AJ, Paulo da Cruz L, Amaro ÉDN, de Araújo Delmondes G, Leite de Oliveira Sobreira Nóbrega JP, Pereira Lopes MJ, Martins da Costa JG, Bezerra Felipe CF, Kerntopf MR (2019). Effects of the *Hyptis martiusii* Benth. leaf essential oil and 1,8-cineole (eucalyptol) on the central nervous system of mice. Food and Chemical Toxicology 133: 110802. doi: 10.1016/j.fct.2019.110802.

Takeda H, Tsuji T, Matsumiya M (1998). Changes in head-dipping behavior in the hole-board test reflect the anxiogenic and/or anxiolytic state in mice *European Journal of Pharmacology* 350: 21-29.

Wei XY, Yang JY, Wang JH, Wu CF (2007). Anxiolytic effect of saponins from *Panax quinquefolium* in mice. *Journal of Ethnopharmacology* 111(3): 613-618. doi: 10.1016/j.jep.2007.01.009.

Yan S, You ZL, Zhao QY, Peng C, He G, Gou XJ, Lin B (2015). Antidepressant-like effects of Sanyuansan in the mouse forced swim test, tail suspension test, and chronic mild stress model. Kaohsiung Journal of Medical Sciences 31(12): 605-612. doi: 10.1016/j.kjms.2015.10.009.

Zhang N, Yao L (2019). Anxiolytic effect of essential oils and their constituents: A review. *Journal of Agricultural and Food Chemistry* 67 (50): 13790-13808. doi: 10.1021/acs.jafc.9b00433.

Zhou Xi-D, Shi D-D, Zhang Z.-J, (2019). Antidepressant and anxiolytic effects of the proprietary Chinese medicine shexiang baoxin pill in mice with chronic unpredictable mild stress. *Journal of Food Drug Analysis* 27: 221-230.

Zimcikova E, Simko J, Karesova I, Remlacek J, Malakova J (2017). Behavioral effects of antiepileptic drugs in rats: Are the effects on mood and behavior detectable in open-field test? *Seizure,* 52: 35-40