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Neurobehavioural Effect of Ethanol Extract of *Adenopus breviflorus* (Roberty) Fruit In Mice

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Abstract

Aim: This study was designed to investigate neurobehavioral effect of Ethanol Extract of *Adenopus breviflorus* fruit (EEAB) in mice.

Methods: Effect of EEAB on the following behavioral activity was studied in mice: novelty-induced rearing and grooming, locomotor activity, head dips and memory. The mechanism of action was studied using the following receptor blockers: atropine, cyproheptadine, haloperidol, naloxone, propranolol and yohimbine. Data were analyzed using descriptive statistics and ANOVA at p=0.05.

Results: The EEAB (250-2000 mg/kg) and diazepam (2.0 mg/kg) caused significant ($p<0.05$) reductions in novelty-induced rearing in mice relative to control. The EEAB (62.5-2000 mg/kg) and diazepam (2.0 mg/kg) caused significant ($p<0.05$) reductions in locomotor activity relative to control. The EEAB (62.5-2000 mg/kg) and diazepam (2.0 mg/kg) caused significant ($p<0.05$) reductions in head dips relative to control. Pretreatment of mice with naloxone, propranolol and yohimbine reversed the decrease in novelty-induced rearing induced by extract (2000 mg/kg) relative to control. Treatment of mice with 1000 mg/kg and 2000 mg/kg of EEAB caused significant ($p<0.05$) increase in percentage alternations relative to control.

Conclusion: It can be concluded that *Adenopus breviflorus* fruit probably has central nervous system depressant effect which could be mediated via μ-opioid, β – adrenergic and α_2 – adrenergic receptors.

Keywords: *Adenopus breviflorus*, Locomotor activity, Memory, Mice, Rearing.

INTRODUCTION

When faced with an unfamiliar environment or object, animals often exhibit behavior patterns that broadly can be termed exploration, such as locomoting around the environment, orientating towards novelty, and touching or sniffing novel objects [1, 2, 3, 4]. Exploration potentially provides an animal with new information about food sources, shelters or mating opportunities. However, by entering a new environment or attending to a novel stimulus, an animal might also increase its risk of predation, aggression from conspecifics or other hazards. Whether an animal investigates or avoids novelty has been described as the outcome of an approach–avoidance conflict [5, 6, 7] or as a balance between neophilic and neophobic tendencies [8]. In motivational terms, neophilia can be defined as the attraction that an animal displays towards an object or place simply because it is novel, while neophobia is the aversion that an animal shows towards approaching a novel object or place [8]. In behavioral terms, neophilia and neophobia can be considered respectively as curiosity-based approach to, and fear-based avoidance of, a novel stimulus [9].

The exploratory behavior of rodents has gained recent interest within a number of areas of behavioral pharmacology. For instance, researchers studying drug addiction are interested in the neural mechanisms underlying neophilia due to the apparent overlap with the neural mechanisms involved in the rewarding effects of drug-taking [10].

Adenopus breviflorus belongs to the family of Cucurbitaceae. It is commonly called Wild colocynth in English language, "Ogbenwa" in Ibo language and "Tagiri" in Yoruba language [11]. It is a perennial tendril climber. It would usually lie on the ground for want of something to climb and climbs over shrubs and herbs by means of axillary tendrils. The leaves are simple, alternate and palmately veined [12].

Medicinally, the plant is used as a purgative in Tanganyika as well as a vermifuge and cathartic in Nigeria [11]. A decoction from the plant is said to be used in Nigeria for headache [11]. It is used in West Africa for a wide range of gastrointestinal disorders and measles in man. In southern Nigeria, its seed-decoction is reportedly given to pregnant women but the purpose is not stated [13]. It is used as an anticonvulsant, sedative and pain killer [14]. It is used with other medicinal plants as concoctions to aid parturition in humans [15]. Livestock farmers employ the fruit extract of the plant for the treatment of Newcastle disease and coccidiosis in animals [15]. The fruit is also used for money-making charms by the Yoruba herbalists of South-Western Nigeria because of the cowrie-like inscriptions on its body.

Pharmacologically, it has been reported that the methanol extract of its whole fruit has anti-implantation activity [16] and abortifacient activity [17]. The ethanol extract of its whole fruit has been reported to have a broad spectrum antibacterial activity [18] as well as anti-oxidant and anti-ulcerogenic effects [19]. Its ethanol extract has been reported to have a little toxic and a lot of beneficial effects on the hematological functions and blood chemistry of male Wistar rats [20].

Since this plant has been reported to have a wide range of neuroactive property [14, 11], this study therefore aims to investigate the neurobehavioural effect of ethanol extract of *Adenopus breviflorus* fruit in mice.

MATERIALS AND METHODS

Experimental Animals

Adult male mice weighing between 20-25 g bred in the Pre-Clinical Animal House of the College of Medicine, University of Ibadan were used. They were housed under standard laboratory conditions and had free access to feed (Ladokun Feeds Limited, Ibadan, Nigeria) and water; they were acclimatized for two weeks to laboratory conditions before the commencement of the experiments. All experiments were carried out in compliance with recommendations of the University of Ibadan Ethics Committee on guiding principles on care and use of animals.

Plant Material

Fresh samples of *Adenopus breviflorus* fruit were bought in Bodija Market, Ibadan, and were authenticated in the Taxonomy Unit of the Forestry Research Institute of Nigeria (FRIN), Jericho, Ibadan where a voucher specimen (FHI 108336) was deposited in their Herbarium.

Preparation of Crude Ethanol Extract

Large quantity (7.5 kg) of fresh specimens of the whole fruit of *Adenopus breviflorus* were washed free of debris and pulverized using mortar and pestle and air-dried for eight weeks. The resultant dried specimens (300 g) were macerated and extracted with 70 % ethanol for 72 hours at room temperature (26 - 28 ^{0}C). The resulting solution was then filtered using a wire-gauze and a sieve with tiny pores (0.25 mm). The 70 % ethanol was later evaporated using steam bath (40 – 45 ^{0}C) to give a percentage yield of 8.6 % of the starting sample. The dried sample was reconstituted in distilled water to make up test solutions of known concentration.

Phytochemical Screening

Standard phytochemical methods were used to test for the presence of alkaloids, cardenolides, tannis, flavonoids, anthraquinones and saponins [21, 22, 23].

(a) Alkaloids test

Few drops of Dragendorff's, Mayer's and Wagner's reagent were added separately to the extract in the test tube. A reddish brown, cream and reddish brown precipitate respectively indicates a positive test.

(b) Keller Killiani's test

The extract was evaporated to dryness and 3 ml of ferric chloride reagent was added to the cooled residue in a clean test tube. Concentrated sulphuric acid (2 ml) was gently poured down the side of the test tube. A purple or reddish brown ring at the interface and green colour in acetic acid layer indicates a positive test for 2-de-oxy sugar (cardenolides' test).

(c) Tannins test

Few drops of ferric chloride reagent were added to the extract in the test tube. A red colouration indicates a positive test.

(d) Flavonoids test

A small quantity of the extract was dissolved in dilute sodium hydroxide and hydrochloric acid was added to the mixture. A yellow solution that turns colorless on addition of hydrochloric acid indicates the presence of flavonoids.

(e) Anthraquinones (Borntrager's) test

Few drops of chloroform were added to the extract in the test tube, 1ml of dilute (10%) ammonial was added and the mixture was shaken. A pink-red color in the ammoniacal (lower) layer shows anthracene derivatives.

(f) Saponins (Froth) test

To 0.5 g of the extract in the test tube was added 10 ml of distilled water and this was well shaken and left to stand for 10 minutes. A thick persistent froth indicated the presence of saponins.

Toxicity test

The method described by [24] was used to determine the LD_{50}, which is the index of acute toxicity. Male albino mice (20-25 g) were used. This method involved an initial dose finding procedure, in which the animals were divided into three groups of three animals per group. Doses of 10 mg/kg, 100 mg/kg and 1000 mg/kg were administered orally, one dose for each group. The treated animals were monitored for twenty-four hours for mortality and general behavior. From the results of the above step, seven different doses (2000 mg/kg, 3000 mg/kg, 4000 mg/kg, 5000 mg/kg, 6000 mg/kg, 7000 mg/kg, 8000 mg/kg) where chosen and administered orally to seven groups of animals of one mouse per group respectively. The treated animals were monitored for twenty-four hours. The LD_{50} was then calculated as the geometric mean of the lowest dose showing death and the highest dose showing no death.

Preparation of Stock Solution of EEAB

Ten grams of EEAB were dissolved in 100 ml of distilled water to give a concentration of 0.1 g/ml.

The dosages of EEAB administered in these studies were obtained from the results of the acute toxicity test.

Behavioral Study

(a) Novelty-induced rearing and grooming

The behavioral profiles of albino mice under the influence of the extract were assessed in the Open Field Box (OFB) (45 cm x 25cm x 25 cm). Forty mice were randomly divided into eight groups (n=5). Group I was given distilled water (0.2 ml/20 g, *p.o.*), groups II – VII were given EEAB (62.5 – 2000 mg/kg, *p.o.*), while group VIII was given diazepam (2.0 mg/kg, i.p.).

Thirty minutes after treatment with the extract behavioral measurements were carried out for a period of thirty minutes. The animals were removed directly from the home cage and placed inside the OFB. Each animal was used only once, with the box cleaned with 70 % ethanol after each assessment to remove olfactory cue from previous animal to the other.

The time of the experiment was kept constant (8.00 a.m.-1.00 p.m.) daily to avoid changes in biologic clock. The behavioral components employed in this observational analysis were rearing and grooming [25]. The frequency of rearing episodes was quantified by using a manual counter and a stop watch. The total frequency was summed up for each animal and totalled for the thirty minutes of observation time.

Rearing was taken as the number of times the mouse was standing on its hind limb or with its forelimbs against the wall of the box or in the free air. Grooming was taken as the number of body cleaning with mouth and face washing with forelimbs.

(b) Locomotor activity

Motor activity was measured in an OFB (45 cm x 25 cm x 25 cm) with painted black grids dividing the floor into 16 (7 cm x 7 cm) equal squares. Forty mice were randomly divided into eight groups (n=5). Group I was given distilled water (0.2 ml/20 g, *p.o.*), groups II – VII were given EEAB (62.5 – 2000 mg/kg, *p.o.*), while group VIII was given diazepam (2.0 mg/kg, i.p.).

One hour after treatment with the extract, each mouse was placed in one of the corners of the box and the numbers of squares crossed with all four paws were counted for 5 minutes. The cage was cleaned with 70 % ethanol at intervals when each animal was removed [26].

(c) Head dips test

The effect of the extract on the rate of head dipping was determined in the hole board which is made up of a number of holes (usually 16) through which the animal can poke its head. Forty mice were randomly divided into eight groups (n=5). Group I was given distilled water (0.2 ml/20 g, *p.o.*), groups II – VII were given EEAB (62.5 – 2000 mg/kg, *p.o.*), while group VIII was given diazepam (2.0 mg/kg, i.p.).

One hour after treatment with the extract, each mouse was placed on the hole board and the number of times that each animal dipped (poked) its head into the holes in 5 minutes were counted [27]. The hole board was cleaned with 70 % ethanol at intervals when each animal was removed.

Mechanism of Action

In another set of experiments, mice were pretreated i.p. for 15 minutes with neurotransmitter blockers to evaluate the mode of action of the extract on novelty-induced rearing and grooming behaviours, locomotor activity and head dips. The following receptor blockers were used: atropine (muscarinic blocker, 0.5 mg/kg), cyproheptadine (5-HT blocker, 0.5 mg/kg), haloperidol (dopaminergic blocker, 0.2 mg/kg), naloxone (μ-opoid antagonist, 2 mg/kg), propranolol (β-adrenergic blocker, 0.2 mg/kg) and yohimbine (α_2-adrenergic blocker, 1.0 mg/kg). The doses administered are the doses that have been found not to induce behavioral effects of their own in experimental animals and as such they only block the receptors involved. The mice were then pretreated for another 30 minutes with maximal dose of the extract (2000 mg/kg). The animals were observed for behavioral responses as previously explained.

(d) Effect on Memory (Y-Maze test)

The Y-maze test can be used as a measure for short term working memory and locomotor activity. Spontaneous alternation is a measure spatial working memory. To alternate among spatial location, a mouse must remember its pervious location. Spontaneous alternation performance was assessed using a Y-maze composed of three equal spaced arms (120^0 38cm x 33cm x 13cm). This test was carried out using this apparatus to obtain results for spontaneous alternation performance (memory) and locomotor activity (total arm enteries). Forty mice were randomly divided into eight groups (n=5). Group I was given distilled water (0.2 ml/20 g, *p.o.*), groups II – VII were given EEAB (62.5 – 2000 mg/kg, *p.o.*), while group VIII was given diazepam (2.0 mg/kg, i.p.).

One hour after pretreatment with the extract, each mouse was placed in one of the arm compartments usually arm A for consistency and was allowed to move freely for 5 minutes. An arm entry is defined as the body of a mouse (except for its tail) completely entering into an arm compartment. The sequence of arm entries is manually recorded. An alternation is defined as an entry into all three arms on consecutive devices. The percentage alternation was expressed as the ratio of actual alternations to possible alternations (defined as the total number of arm entries minus two) multiplied by 100. Ethanol (70 %) was used to clean the Y-maze at interval when each animal was removed [28].

Statistical Analysis

The mean and standard error of mean (S.E.M) were calculated for all values. Comparison between the control and experimental groups was done using one - way analysis of variance (ANOVA) with Duncan's Multiple Range Test. Differences were considered statistically significant at $p<0.05$.

RESULTS

Phytochemical analyses of the crude extract revealed the presence of alkaloid, cardenolides, tannins and flavonoids, while anthraquinones and saponins were absent (Table 1).

The LD_{50} of the crude extract was found to be 7000 mg/kg *p.o.*

The administration of EEAB (250 mg/kg, 500 mg/kg, 1000 mg/kg, 2000 mg/kg) and diazepam (2.0 mg/kg) to mice caused significant ($p<0.05$) reductions in novelty-induced rearing relative to the control (Figure 1). Treatment of mice with all the treatment doses of EEAB (62.5 mg/kg, 125 mg/kg, 250 mg/kg, 500 mg/kg, 1000 mg/kg, 2000 mg/kg) and diazepam (2.0 mg/kg) caused significant ($p<0.05$) reductions in grooming relative to the control (Figure 2). Also, treatment of mice with all the treatment doses of the extract (62.5 mg/kg, 125 mg/kg, 250 mg/kg, 500 mg/kg, 1000 mg/kg, 2000 mg/kg) and diazepam (2.0 mg/kg) caused significant ($p<0.05$) reductions in locomotor activity relative to the control (Figure 3). Treatment of mice with all the treatment doses of EEAB (62.5 mg/kg, 125 mg/kg, 250 mg/kg, 500 mg/kg, 1000 mg/kg, 2000 mg/kg) and diazepam (2.0 mg/kg) caused significant ($p<0.05$) reductions in head dips relative to the control (Figure 4).

Pretreatment of mice with atropine, cyproheptadine and haloperidol did not reverse the decrease in novelty-induced rearing, grooming and locomotor activity induced by EEAB (2000 mg/kg) relative to the control; these antagonists potentiated the decrease in novelty-induced

rearing and grooming induced by the extract. Pretreatment of mice with naloxone, propranolol and yohimbine reversed the decrease in novelty-induced rearing induced by the extract (2000 mg/kg) relative to the control, but did not reverse the decrease in novelty-induced grooming and locomotor activity induced by EEAB (2000 mg/kg) relative to the control (Table 2). Pretreatment of mice with atropine, cyproheptadine, haloperidol, naloxone, propranolol and yohimbine did not reverse the decrease in head dips induced by EEAB (2000 mg/kg) relative to the control (Table 3).

Treatment of mice with 1000 mg/kg and 2000 mg/kg of EEAB caused significant ($p<0.05$) increase in percentage alternation relative to the control, while diazepam (2.0 mg/kg) caused significant decrease in percentage alternation relative to the control (Figure 5).

Table 1: Phytochemical constituents of EEAB

Phytochemicals	EEAB
Alkaloids	+
Cardenolides	+
Tannins	+
Flavonoids	+
Anthraquinones	-
Saponins	-

+ = Present
- = Absent

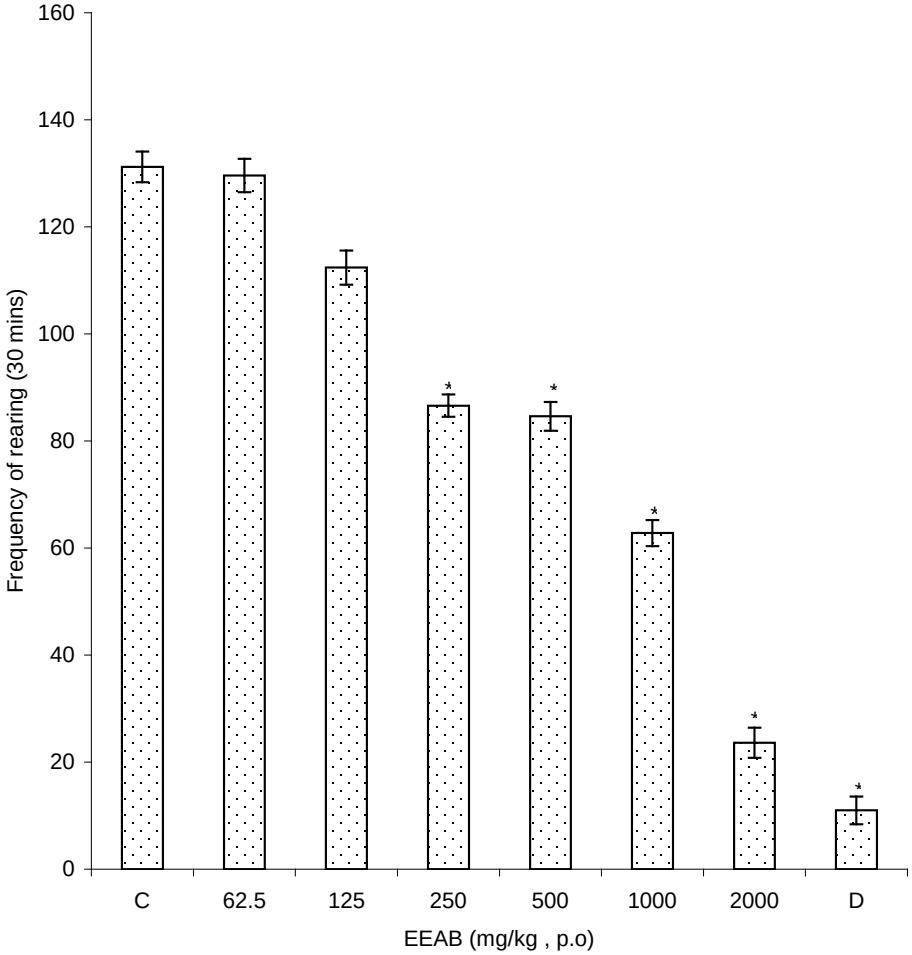

Figure 1: Effect of EEAB on novelty-induced rearing in mice
C: Control, D: Diazepam (2.0 mg/kg, i.p.)

The results are expressed as mean values ± S.E.M. (n=8). One way ANOVA revealed significant difference [$F(7, 32) = 28.784$, $p<0.05$] between various treatment groups. * Indicates significant difference from control at $p<0.05$.

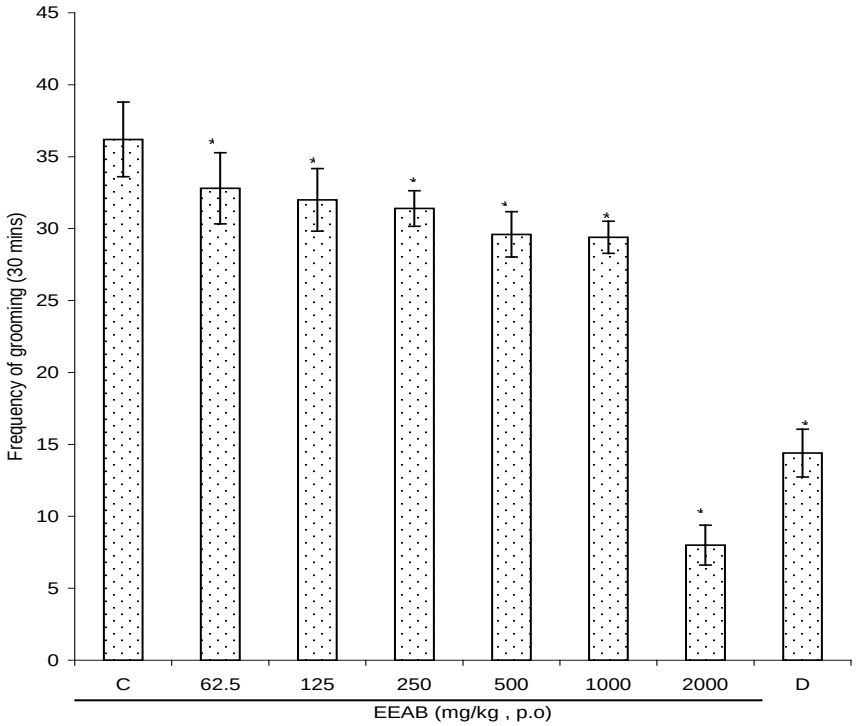

Figure 2: Effect of EEAB on novelty-induced grooming in mice
C: Control, D: Diazepam (2.0 mg/kg, i.p.)

The results are expressed as mean values ± S.E.M. (n=8). One way ANOVA revealed significant difference [F (7, 32) =12.551, p<0.05] between various treatment groups. * Indicates significant difference from control at p<0.05.

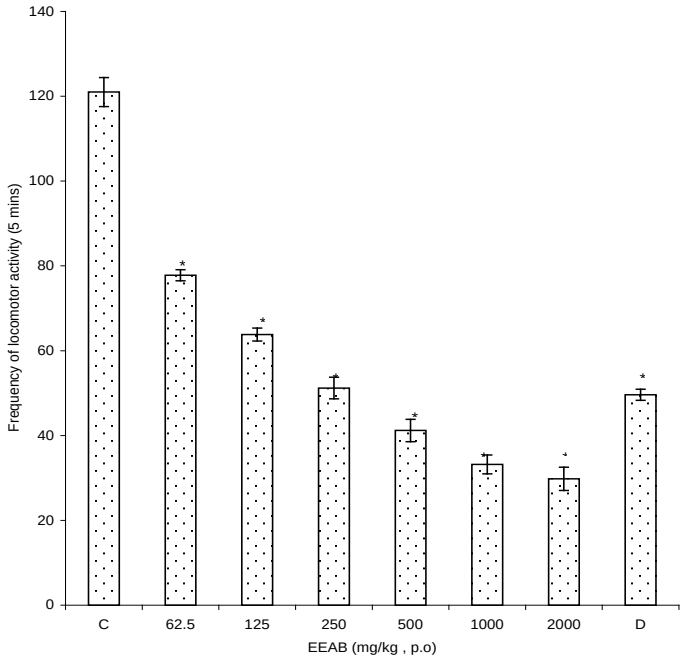

Figure 3: Effect of EEAB on locomotor activity in mice C: Control, D: Diazepam (2.0 mg/kg, i.p.)

The results are expressed as mean values ± S.E.M. (n=8). One way ANOVA revealed significant difference [F (6, 28) =9.347, p<0.05] between various treatment groups. * Indicates significant difference from control at p<0.05.

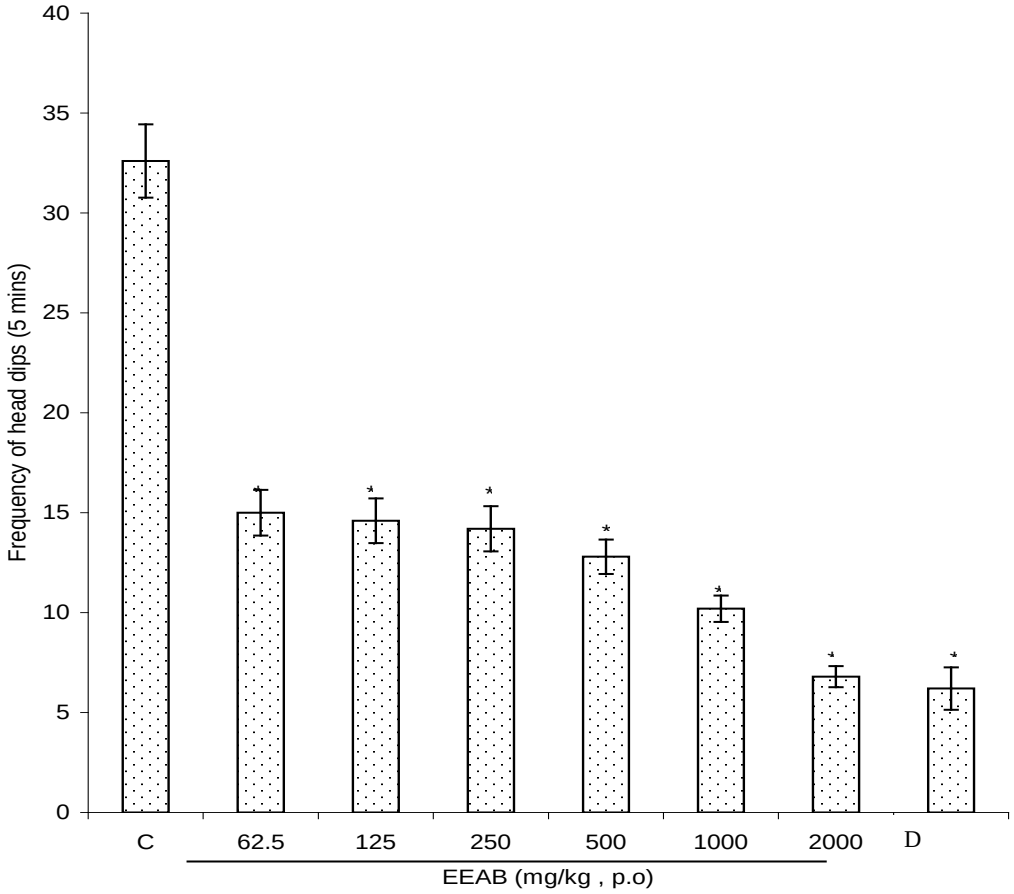

Figure 4: Effect of EEAB on head dips in mice

C: Control, D: Diazepam (2.0 mg/kg, i.p.)

The results are expressed as mean values ± S.E.M. (n=8). One way ANOVA revealed significant difference [F (7, 32) =40.561, p<0.05] between various treatment groups. * Indicates significant difference from control at p<0.05.

Table 2: Effect of EEAB on novelty-induced rearing, grooming and locomotor activity in presence of antagonists

Treatment	Dose (mg/kg)	NIR/30 min	NIG/30 min	LA/ 5 min
Control	0.2ml/20g	131.20 ± 2.86	36.20± 2.59	121.00 ± 3.42
EEAB	2000	23.60 ± 2.82*	8.00 ± 1.39*	29.80 ± 1.82*
Atropine	0.5	119.80 ± 2.66	32.60 ± 2.61*	93.20 ± 0.92*
Atropine +EEAB		4.20 ± 0.62*	7.80 ± 1.35*	31.40 ± 1.27*
Cyproheptadine	0.5	109.80 ± 3.21	24.00 ± 1.09*	94.00 ± 2.51*
Cyproheptadine + EEAB		4.80 ± 0.83*	2.60 ± 0.40*	27.20 ± 1.66*
Haloperidol	0.2	101.80 ± 2.54*	24.60 ± 0.65*	66.00 ± 2.86*
Haloperidol + EEAB		19.80 ± 1.71*	6.00 ± 0.75*	30.80 ± 2.01*
Naloxone	2.0	114.60 ± 2.63	15.60 ± 0.95*	63.60 ± 1.72*
Naloxone + EEAB		112.40 ± 2.92	10.80 ± 0.82*	42.20 ± 2.68*
Propranolol	0.2	120.00 ± 2.61	26.80 ± 2.01*	60.80 ± 2.28*
Propranolol + EEAB		113.80 ± 2.77	23.80 ± 0.82*	59.00 ± 2.72*
Yohimbine	1.0	112.8 ± 1.91	29.40 ± 1.43*	67.20 ± 2.68*
Yohimbine + EEAB		110.40 ± 1.32	24.40 ± 1.69*	59.00 ± 2.07*

The results are expresses as mean ± S.E.M. (n=8). One way ANOVA revealed significant difference between various treatment groups. * Indicates significant difference from control at *p<0.05.

NIR: Novelty induced rearing, NIG: Novelty induced grooming, LA: Locomotor activity.

Table 3: Effect of EEAB on head dips in presence of antagonists

Treatment	Dose (mg/kg)	HD/ 5 min
Control	0.2ml/20g	32.60 ± 1.83
EEAB	2000	6.80 ± 0.53*
Atropine	0.5	20.20±1.20*
Atropine +EEAB		7.40 ± 0.57*
Cyproheptadine	0.5	10.80±0.63*
Cyproheptadine + EEAB		3.40 ± 0.60*
Haloperidol	0.2	10.00±0.79*
Haloperidol + EEAB		4.00 ± 0.53*
Naloxone	2.0	10.20±0.37*
Naloxone + EEAB		4.20 ± 0.53*
Propranolol	0.2	9.80 ± 0.39*
Propranolol + EEAB		9.80 ± 0.55*
Yohimbine	1.0	9.80 ± 0.42*
Yohimbine + EEAB		8.20 ± 0.37*

The results are expresses as mean ± S.E.M. (n=8). One way ANOVA revealed significant difference between various treatment groups. * Indicates significant difference from control at *p<0.05.

HD: Head dip

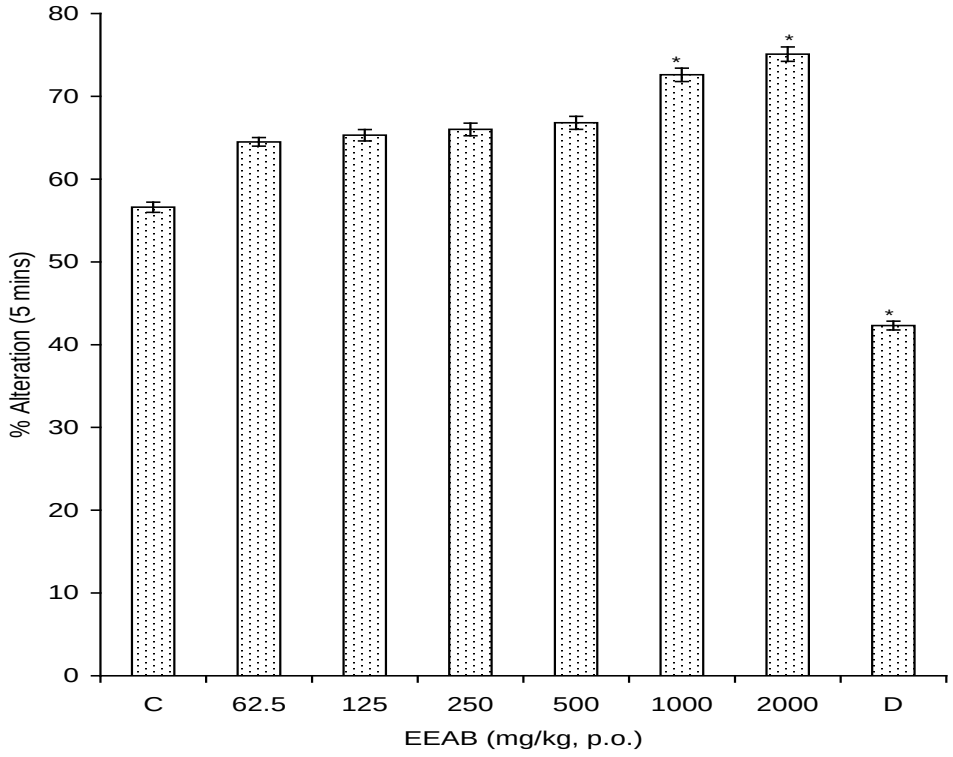

Figure 5: Effect of EEAB on memory in mice (% Alternation)
C: Control, D: Diazepam (2.0 mg/kg, i.p.)

The results are expressed as mean values ± S.E.M. (n=8). One way ANOVA revealed significant difference [F (7, 32) =5.014, p<0.05] between various treatment groups. * Indicates significant difference from control at p<0.05.

DISCUSSION

Phytochemical analysis is an important tool to identify and screen plants for their use [29]. The phytochemical analysis of the extract indicated the presence of tannins, and it has been reported that herbs that have tannins as their major components are astringent in nature and are used for treating intestinal disorders such as diarrhea and dysentery [30]. Therefore, these observations could support the use of this plant in West Africa for treating a wide range of gastrointestinal disorders [14]. The analyses also revealed the presence of alkaloids and flavonoids which have been reported to possess various important pharmacological activities including inflammatory, antioxidant and antinociceptive activities [31, 32] which justify the use of this plant in herbal cure remedies.

Acute toxicity test gives clues on the range of doses that could be toxic to the animal; it could also be used to estimate the therapeutic index (LD_{50}/ED_{50}) of drugs and xenobiotics

[33]. LD_{50} is the dose at which mortality occurs in 50% population of the experimental animals. The higher the value of the LD_{50} for a substance, the relatively safer the substance is assumed to be. The LD_{50} determination for the extract in mice via the oral route was 7000 mg/kg, which was not toxic to the animals and since the recommended single high dose by OECD guidelines 423 [34] for testing acute toxicity is 2000 mg/kg; this probably indicates the extract has wide safety margins (low toxicity). Similar result was reported by [35] in *Eichhornia crassipes* extract treated mice.

The extract was examined for novelty – induced rearing (NIR) in mice. The NIR is a behavior of rodents in novel environments. The behavior is employed by rodents as one of the survival strategies in assessing the environment for food, protection and possibly escapes [36]. Measurement of the frequency of rearing in rodents and the modification can therefore be employed in assessing test drugs and extracts for both sedative property and central nervous system stimulation [37]. Rearing has been described as the vertical locomotion activity when the animal stands on its hind leg while raising up its forearm in the air or placed on the wall of the cage [38]. Drugs that stimulate the CNS increase rearing behavior, while those that depress the CNS inhibit rearing behavior. The extract inhibited NIR in mice which probably indicate a sedative or depressant property. Similar result was reported by [39] in Nigerian Honey treated mice.

The crude extract was examined for novelty – induced grooming (NIG) in mice. Grooming is an important behavioral component in animals and is associated with de-arousal of the central nervous system (CNS). De – arousal indicates absence of stimulation. Grooming is described in animals (rat or mice) as face or head washing with forearm or body grooming with mouth [40]. Drugs that have depressant effect inhibit grooming behavior. The extract reduced NIG in mice which suggest that the extract have depressant effect on the CNS. Similar result was reported by [41] in *Alpinia zerumbet* essential oil treated mice.

Locomotor activity is considered as an index of alertness and a reduction is indicative of sedative activity [42]. The open field test is a simple assessment used to establish the general activity levels, gross locomotor activity and exploration habits of rodents [43]. The extract caused reduction in locomotor activity which further confirms the CNS depressant activity of the extract. Similar result was reported by [44] in *Tecoma stans* flowers extracts treated mice.

The hole board is a method used to measure the animal's response to a novel environment and to assess emotionality, anxiety and/or responses to stress [45]. In this test, head dipping behavior may change in response to the emotional state of the animal and an increase in this behavior could reflect the expression of an anxiolytic reaction of the animal [46]. On the other hand, a decrease in the number of head dipping reveals a sedative or depressant behavior [47, 48]. Since, the extract caused decrease in head dips, this probably indicates that the extract possess a sedative or CNS depressant property. Similar result was reported by [49] in *Dracocephalum moldavica* extract treated mice.

Novelty – induced rearing and grooming behavioral responses is regulated by multiple neurotransmitter system, such transmitters include gamma – aminobutyric acid (GABA), cholinergic, adrenergic, opioid, serotonin, glutamate and dopamine receptors [50]. The administration of atropine, cyproheptadine and haloperidol to mice did not reverse the inhibitory effect of the extract on novelty – induced rearing, grooming, head dips and locomotor activity; this probably indicates that muscarinic, serotonergic and dopaminergic receptors were not involved in the inhibitory effect of the extract on the aforementioned behavioral responses. However, the administration of naloxone, propanolol and yohimbine to mice reversed the inhibitory effect of the extract on novelty – induced rearing, this suggests the involvements of μ - opioid, β-adrenergic and α_2 – adrenergic receptors in the inhibitory effect of the extract on the aforementioned behavioral response.

The Y – maze has been reported to be used as a measure of short term memory, general locomotor activity and stereotypical behavior [51, 52]. Learning and memory is one of the most important functions of the brain, which is associated with complex neurophysiologic and neurochemical changes. Many neurotransmitters including acetylcholine, dopamine, norepinephrine and serotonin play an important role in the learning and memory processes [53]. It is well known that spontaneous alternation is a measure of spatial working memory and to alternate among spatial locations, a rodent (rat or mouse) must remember its previous location. The extract induced increase in percentage alternation which probably indicates an enhancement of spatial working memory. Similar result was reported by [54] in *Parkia biglobossa* extract treated rats.

CONCLUSION

It can be concluded that *Adenopus breviflorus* fruit probably has central nervous system depressant effect which provides scientific bases to the folkloric claims of the plant as a neuroactive herbal remedy. Its central nervous depressant effect could be mediated via μ-opioid, β – adrenergic and α_2 – adrenergic receptors.

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