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# Ibie Leave (*Mucuna flagellipes*) Decreased Anxiety Like Behaviour in Swiss White Mice Following Long Term Consumption

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## **Abstract**

The use of *Mucuna flagellipes* in recent time has been on the increase as a result of its speculated therapeutic effect. Owing to the fact that there are no detailed reports on *Mucuna flagellipes* or any report on its effect on anxiety and fear, this work is therefore intended to investigate the possible effects of this herb on anxiety and fear in Swiss white mice, using the elevated plus maze as experimental apparatus. Twenty CD-1 mice were divided into two groups (1-2, n=10). Group 1 (Control) was administered normal fed while group 2 was the treated group. Data were analyzed using statistical tools (one way ANOVA and Student t-test). Results revealed anxiety like behavior treated mices when compared to the control. However, administration of the Ibie Leave (*Mucuna flagellipes*) reduced anxiety related behavior in mice. In summary, it is therefore a strong consideration that *Mucuna flagellipes* (Ibie leaf) contains compounds and other constituents which could be responsible for the anxiolytic properties and effects which the leaf had on the mice thereby reducing fear and anxiety related behavior.

Keywords: Mucuna flagellipes; Mice; Medicines; Protein; Fat

#### Introduction

Generally, the use of herbs for medicinal purposes is believed to have been passed to generations through oral tradition. Some of this information has been complimented with diverse experiences gained through intercultural exchange and migration. This has led to more curative conditions consequently; as man has relied. The world Health Organization (WHO) estimates that 80% of the world's population relies on these alternative plant based remedies as their primary medical intervention. In as much as medicinal herbs play major roles in the prevention and treatment of diseases, and serves as catalyst in drug development, we do not therefore assure of its safety for uncontrolled use by the public [1]. It is just like any other therapeutic agent, when overdosed or miss-used, has the ability to cause adverse effects. Because herbal medicines are considered safe and nontoxic, the toxicological actions of these herbs have been largely ignored even when its effectiveness is either known or under study [2,3]. The likelihood of adverse effects increases when production and consumption are largely uncontrolled and consumer not adequately informed [4,5].

Anxiety is an emotion characterized by an unpleasant state of inner turmoil, often accompanied by nervous behavior. In this context, while fear is a response to a real or perceived immediate threat, anxiety responses originate from the misinterpretation of potential danger or even expectation of future threat. In some individuals, however these anxiety responses may become persistent, uncontrollable excessive and inappropriate.

The seeds have been reported to be a good source of protein and fat [6-8]. There is abundance of macro elements like calcium, magnesium and iron [6,9]. The pod hairs have been used in India to treat snakebite

[10], while in West Africa, it is the stem that is used for treating of snakebite also [11]. It stem have been reported to also improve sexual potency [9,12]. pods have also been used to deworm patients [13]. The modus operandi of the pod hair is mechanical, thus when mixed with syrup, molasses and honey, pierces the bodies of the intestinal worms, twisting themselves free from the walls so that a brisk catharsis will push them out. Many researchers tend to overlook this plant, been justified by less literatures found on the plant. This could be attributed to its itching brown hairs on the S-shaped fruit which discourage not only man but animals. Therefore, the aim of the study was to investigate the effects of the plant anxiety and fear in Swiss white mice so as to use the leaves of this plant for the management/treatment of neurobehavioral disorders such as anxiety disorders if the leaf of plant is found to have a positive effect on this neurobehavioral parameter.

# **Materials and Methods**

Dried leaves of Mucuna flagellipes was obtained from a local market in Okigwe, Abia State, Nigeria. The leave was authenticated in the Herbarium unit of the Department of Biochemistry, Faculty of Sciences, Abia State, Nigeria.

# **Experimental animals**

Experimental animals (male and female mice) between 21-35 days old and weighing between 19-21 g were obtained from the animal house of the Department of Human Physiology, Faculty of Medicine, Abia State University and housed in a new wired cages in the same animal house were the mice acclimatized for two weeks. Prior to the commencement of the experiments. The mice were housed under standard laboratory condition, light and dark cycles of 12 h and were provided standard rodent pellet diet and water ad libitum. The mice were categorized into control and treated groups. The treated group

was administered, in addition to feed and water for a period of three weeks.

# Neurobehavioral set-up

The set-up used for the neurobehavioral studies was the Elevated plus maze apparatus.

# Elevated plus maze

The EPM set-up was built according to the description of Lister [14] as reported by John [15]. The apparatus is in the configuration of a '+' and comprised of two open arms (50  $\times$  10 cm) across from each other and perpendicular to two closed arms (50  $\times$  10  $\times$  20 cm) with a central platform (10  $\times$  10 cm). The closed arms had a high (20 cm) wall to enclose the arm. The entire apparatus was made of wooden materials, painted black.

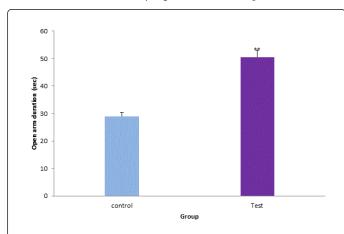
#### **Experimental procedure**

Mice were placed in the central square of the plus maze facing an open arm and then allowed to explore the apparatus for 5 minutes. The maze was then cleaned with methylated spirit and allowed to dry between tests. Behaviors scored were: Open arm duration, close arm duration, head dipping, grooming, rearing, etc.

#### Results

#### Behaviors scored using the elevated plus maze (EPM)

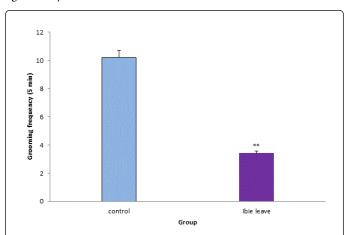
**Open arm duration:** Figure 1 compares the open arm duration in the two groups of mice. The anxiety behavior following administration of Ibie leave and control diet was measured by the duration of time the animals (mice) spent in the open arm within five minutes in the elevated plus maze. The values were,  $29.00 \pm 3.49$  (control) and  $50.51 \pm 6.22$  (Ibie leave). Figure 1, shows that the open arm duration of the Ibie leave fed mice was statistically higher (P<0.01) compared to control.



**Figure 1:** Comparison of Open Arm Duration in the elevated plus maze in the control and Ibie leave treated groups. Values are mean  $\pm$  SEM, n=9. \*\*p<0.01 vs. control.

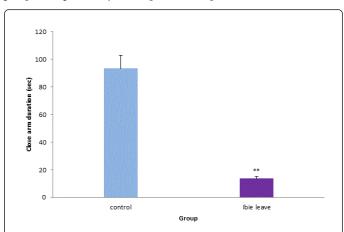
**Grooming frequency:** The frequency of grooming in the EPM for control mice and test group was  $10.20 \pm 2.13$  and  $3.40 \pm 0.74/5$  min (Ibie leave) respectively. The graph in Figure 2 shows that the

frequency of grooming among the groups, when compared was significantly different at P<0.01.



**Figure 2:** Comparison of Grooming Frequency in the elevated plus maze in the control and Ibie leave treated groups. Values are mean  $\pm$  SEM, n=9. \*\*p<0.01 vs. control.

**Close arm duration:** Figure 3 compares close arm duration in the two experimental groups. The values are:  $93.40 \pm 13.74$  (control) and  $51.20 \pm 3.48$  (Ibie leave). The duration of the close arm of the Ibie leave group was significantly lower (p<0.01) compared to control.



**Figure 3:** Comparison of Closed Arm duration in the elevated plus maze in the control and Ibie leave treated groups. Values are mean  $\pm$  SEM, n=9. \*\*p<0.01 vs. control.

**Stretch attend posture:** The frequency of stretch attend posture between the two experimental groups is in Figure 4. The values are:  $5.40 \pm 0.87$  (control) and  $2.40 \pm 0.24$  (Ibie leave). The frequency of SAP of the group of mice fed with Ibie leave was statistically lower (p<0.001) compared to control.

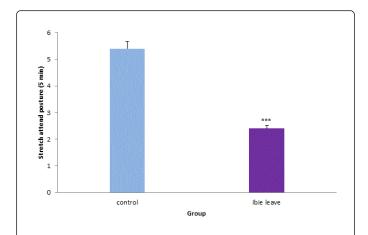
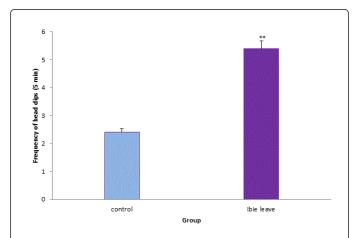


Figure 4: Comparison of Stretch Attend Posture in the open field maze in control and Ibie Leave-treated groups. Values are mean ± SEM, n=9. \*\*p<0.001 vs. control.

Frequency of head dips: The frequency of head dips between the mice fed with Ibie leave and control are: 2.40  $\pm$  0.24 min and 5.40  $\pm$ 0.77 min. The frequency of head dips for the group of mice fed with Ibie leave was significantly higher (0.01) compared to control (Figure



**Figure 5:** Comparison of frequency of Head dips in the elevated plus maze in the control and Ibie leave treated groups. Values are mean  $\pm$  SEM, n=9. \*\*p<0.01 vs. control.

**Defecation frequency:** The frequency of defecation between the two experimental groups is in Figure 6. The values are:  $0.40 \pm 0.24$  (control) and  $0.20 \pm 0.20$  (Ibie leave). The frequency of defecation of the group of mice fed with Ibie leave was not significantly different compared to control.

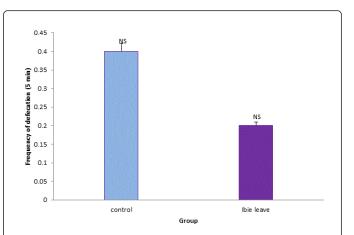


Figure 6: Comparison of Defecation Frequency in the open field maze in the control and Ibie Leave-treated groups. Values are mean ± SEM, n=9. NS=Not Significant.

#### Discussion

The Elevated Plus Maze is widely used behavioral assay for rodents and it has been validated to assess the anti-anxiety effects and to define brain regions and mechanisms underlying anxiety-related behavior [16,17]. It is a validating and reliable test for detecting both anxiolytic and anxiogenic-like effects of agents [18,19]. In this study, specific variables, such as time spent in the open/close arm entry frequencies to the open/close arm, grooming, SAP and head dipping were used to characterize the level of anxiety in mice. Elevated anxiety levels observed as increased entry frequency into the close arm; increased grooming and head dipping frequencies in the Ibie leave treated group implies enxiogenic effect of Mucun flagellipes. This finding is in line with previous studies related to the Ibie leave, such as chronic consumption of Ibie leave on locomotor behavior in CD-1 mice [20] and the ethanolic extract of the leave on memory and learning in mice [21]. An increase in time and proportion or duration of the entries into the open arms without a changed locomotor activity is regarded as a powerful marker for an anxiolytic substance effect [22]. In this study, administration of Ibie leave remarkably decreased anxiety levels by reduced entry or duration/frequency into the close arm across the period of the study when compared to control. Anxiogenic response, such as, grooming, SAP and head dipping, grooming has been demonstrated as credible indices in measuring rodent response to stress [16]. Striking decrease in anxiety-like response, such as, grooming and head dipping frequencies were observed with Ibie leave treatments. Increase in the duration and frequencies of the entries into the open arms in EPM test had been confirmed as a potent sign of an anxiolytic substance effect [18].

Several researchers have reported that flavonoid groups exhibited a wide range of biological activities, such as antioxidant, antiinflammatory, and antiangiogenic effects [23]. It is possible that phytochemical components with antioxidant activities such as flavonoid play essential roles in the anxiolytic properties of Ibie leave as observed in this study. This is in consistence with report related to anxiolytic effect of the plant extracts [24].

## Conclusion

The Ibie leave tends to have a significant effect on the nervous system by reducing the level of anxiety in animals. The plant also has low toxicity and reduced side effects, therefore pharmaceutical industries can use this plant in producing safe and efficient drugs for people with anxiety disorders. Traditional medicine practitioners can also use this plant in making herbal mixtures for the treatment of anxiety disorders in rural areas, making it available and cheap for people living in those areas. Therefore, since the Ibie leave may prove efficacious in ameliorating anxiety-like behavior in mice. It would be worthwhile to explore the potential of this plant in the management/ treatment of anxiety disorders.

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