

## **Behaviour of male Wistar rats in response to different doses of extract of *Isoberlinia doka***

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### **ABSTRACT**

*The present study investigated the effects of different doses of stem-bark extracts of *Isoberlinia doka* (I.D) on neuromuscular and sensorimotor responses of 35 male Wistar rats. The results of the study showed that oral administration of the extract at 200 and 600mg/Kg body weight did not elicit any significant ( $P > 0.05$ ) change in neuromuscular and sensorimotor responses of male Wistar rats. However, the administration of the extract at doses of 1000, 1400, 1800 and 2200mg/Kg body weight significantly ( $P < 0.05$ ) decreased the neuromuscular and sensorimotor responses of the rats. In conclusion, the present study, for the first time, showed that higher doses of I.D above 600mg/kg body weight inhibited neuromuscular and sensorimotor pathways, which have resulted to the impairment of postural stability, skill performance, motor strength, placing and righting reflex responses. Thus, the administration of extract of I.D by individuals should be done with great caution.*

**Keywords:** Extract, *Isoberlinia doka*, male rats, neuromuscular, sensorimotor.

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### **INTRODUCTION**

World Health Organization estimated that 80 % of people world wide rely on herbal medicines for some aspect of their primary health care [1]. This is true, especially in developing countries where primary health care system is inadequate and crude extract from different plants have been used to attend to the health care need of people [2].

Substantial research has been done on many plants that have medicinal properties [3, 4], yet there remain a large number of very important medicinal plants that are yet to be investigated using the current scientific knowledge.

The plant *Isobertinia doka* (I.D) belongs to the Family Caesalpinioideae and is found in the Sudanese and Guinean Savannahs, on well drained clay and average soils, and distributed from Guinea to Cameroon, as far as Sudan. It is a plant of economic value in the wood industry. The roots of I.D are used against nausea, hepatitis, the bark as a vermifuge and healing, medico-religious uses (against curses) while the stem and leaves are used in combating convulsion [5]. The plant has also been shown to alleviate erectile dysfunction [6], traditional treatment of typhoid fever [1], scrotal elephantiasis, infertility and jaundice [4] and as antivenom [7]. To date scientific study on the medicinal property of I.D plant is limited or lacking in the available literature.

The present study investigated the effects of graded doses of water extract of stem bark of I.D on neuromuscular and sensorimotor responses of male Wistar rats.

## MATERIALS AND METHODS

### **Plant and preparation of aqueous extract of *Isobertinia doka***

The plant parts used in this study were collected from the rangeland of the College of Agriculture and Animal Science, Ahmadu Bello University, Mando, Kaduna. The plant part was identified as I.D at the Herbarium of the Department of Biological Sciences, Ahmadu Bello University, Zaria, Nigeria, where the voucher specimen is deposited.

The stem bark of I.D were chopped into pieces, air dried at room temperature, and then pulverized using a Kenwood® blender. A sample (600g) of the powdered stem bark was boiled in 3000 ml of distilled water for an hour. It was then filtered hot through a muslin cloth before further filtration using a Whatman No. 1 filter paper. At the end of the extraction, the filtrate was concentrated in a hot air oven at 50°C and subsequently air dried. The extract was pounded into powder using a porcelain pestle and mortar and then stored in an air tight container and kept at 4°C till when required.

### **Phytochemical screening**

The aqueous stem bark extract was screened as described by Harborne [8]. The stem-bark extract was found to contained secondary metabolites such as flavonoids(++), alkaloids (++) , tannins (+), glycosides (++) , saponin (+) and volatile oil (+).

### **Experimental animals**

A total of 35 male Wistar rats weighing between 113.3 – 192.6g were used for the study. They were purchased from the Department of Pharmacology and Therapeutics, Faculty of Pharmaceutical Sciences, Ahmadu Bello University, Zaria, Nigeria. They were transported to the College laboratory and maintained in clean rat cages in a 12h light/dark cycle with the litter changed every week. They were fed pelletized commercial rat feed (ECWA® Nigeria PLc, Jos, Nigeria) and water was provided *ad libitum* and allowed to acclimatize for a week. The rats were strictly handled and maintained in accordance with the internationally accepted principles for laboratory animal care and use [9]

**Administration of aqueous extract of *Isoberlinia doka***

The male rats were randomly divided into 6 groups of 5 rats each and were administered orally and individually the extract of I.D diluted in sterile water at the dosage rate of 200, 600, 1000, 1400, 1800, 2200mg/Kg body weight, respectively on day 1, 7, 14 and 21 of the study period. The control group was given 1ml of sterile water orally.

**Behavioral Tests***Postural reflex*

Postural reflex was determined as earlier described by Abou-Donia *et al.* [10]. Briefly, the rats were gently held by the tail a meter above the floor and extension of fore limb were observed and graded as follows:

*Grade 0 – No consistent flexion of the fore limbs observed*

*Grade 1 – Moderate consistence flexion was observed*

*Grade 2 – Excessive fore limb flexion.*

*Surface placing reflex*

This behaviour was assessed as earlier described [10]. Rats were gently held by the tail suspended toward the edge of a table. Care was taken to avoid the vibrissae from touching the table. The response of the rats fore limb in contact with the edge of the table was scored as follows:

*Grade 0 – There was immediate placing of the fore limbs to the edge of the table*

*Grade 1 – Slow placing response.*

*Grade 2 – No placing response for a period of 2 – 3 sec.*

*Motor strength (grip time)*

This behaviour was assessed by hanging down a rat from a dowel gripped by the rat. The time taken for the rat to loss it grip and fall down was recorded in seconds [10].

*Surface righting reflex*

For the assessment of this behaviour a rat was placed on a flat smooth leveled table. The rat was then held by the base of the tail and is quickly turned clockwise from a prone to a supine position. The time taken for the rat to return to it normal position was graded as follows [10].

*Grade 0 – Rat that righted itself within 2 sec.*

*Grade 1 – Rat that righted itself after 3-5 sec, which signified impairment of motor coordination.*

*Grade 2 – Rat that righted itself after 5 sec.*

**Statistical Analysis**

Data were analyzed by 1-way analysis of variance (ANOVA) followed by Tukey's test.

**RESULTS****Surface righting reflex**

The result of surface righting reflex showed that all the rats that were administered 1400, 1800 and 2200mg of the extracts had righting reflex of 1.3 to 1.7, which was higher ( $P < 0.05$ ) than those of the control group and rats that were treated with 200, 600 and 1000mg of the extracts

which had righting reflex between 0 to 0.5. The righting reflex was not affected by the period of the administration of the extracts (Table 1).

**Table 1: Surface righting reflex of male Wistar rats in response to different doses of extracts of *Isoberlinia doka* (for each group n = 5).**

Days	Dose (mg/kg body weight)						
	Control	200	600	1000	1400	1800	2200
1	0.8±0.01	0.2±0.01	0	0.2±0.01	1.0±0.01	1.2±0.07	1.6±0.51
7	0	0	0	0.4±0.01	1.5±0.05	1.8±0.11	1.8±0.50
14	0.2±0.01	0.6±0.02	0.6±0.05	0.6±0.05	2.0±0.01	2.1±0.50	1.6±0.25
21	0.2±0.01	0.4±0.05	0.4±0.04	0.8±0.02	0.5±0.01	1.2±0.11	1.6±0.25
Mean ± SEM	0.3±0.01 <sup>a</sup>	0.3±0.01 <sup>a</sup>	0.3±0.11 <sup>a</sup>	0.5±0.01 <sup>a</sup>	1.3±0.20 <sup>b</sup>	1.6±0.12 <sup>b</sup>	1.7±0.10 <sup>b</sup>

Overall mean values with different superscript alphabets along the same row are significantly different ( $P < 0.05$ ).

### Postural reflex

The result of postural reflex in groups 1, 2 and control was 0 to 0.4 rating throughout the study period. In group 3, 4, 5 and 6 rats the postural reflex was between 0.8 to 1.4 ratings. In the overall the percent number of rats with higher rating was significantly higher ( $P < 0.05$ ) in groups 3, 4, 5 and 6 compared to control and group 1 and 2 rats (Table 2).

**Table 2: Postural reflex of male Wistar rats in response to different doses of extracts of *Isoberlinia doka* (for each group n = 5).**

Days	Dose (mg/kg body weight)						
	Control	200	600	1000	1400	1800	2200
1	0	0.2±0.01	0.6±0.01	0.4±0.07	0.8±0.07	0.6±0.05	0.6±0.01
7	0	0	0	1.4±0.51	1.5±0.08	2.0±0.41	1.8±0.45
14	0	0	0.8±0.05	0.6±0.05	0.8±0.01	2.0±0.51	0.6±0.01
21	0	0	0	0.8±0.05	0.5±0.01	0.8±0.01	1.0±0.01
Mean ± SEM	0 <sup>a</sup>	0.1±0.01 <sup>a</sup>	0.4±0.11 <sup>a</sup>	0.8±0.50 <sup>b</sup>	0.9±0.50 <sup>b</sup>	1.4±0.45 <sup>b</sup>	1.0±0.30 <sup>b</sup>

Overall mean values with different superscript alphabets along the same row are significantly different ( $P < 0.05$ ).

### Tactile placing response

The tactile placing response values of 0.7 to 1 recorded in rats administered 1400, 1800 and 2200mg of the extract was significantly ( $P < 0.05$ ) higher than the values of 0.2 to 0.4 recorded in control and group 1, 2 and 3 rats (Table 3).

**Table 3: Tactile placing reflex of male Wistar rats in response to different doses of extracts of *Isoberlinia doka* (for each group n = 5).**

Days	Dose (mg/kg body weight)						
	Control	200	600	1000	1400	1800	2200
1	0.6±0.01	0.2±0.02	0	0	0	0	1.5±0.05
7	0	0.2±0.01	0	0.8±0.05	1.8±0.02	1.4±0.01	1.0±0.01
14	0	0.2±0.05	0.8±0.01	0.4±0.03	1.0±0.11	2.5±0.50	0.7±0.03
21	0	0	0.4±0.01	0.4±0.01	0	0.4±0.01	1.0±0.01
Mean ± SEM	0.2±0.0 <sup>a</sup>	0.2±0.01 <sup>a</sup>	0.3±0.10 <sup>a</sup>	0.4±0.30 <sup>a</sup>	0.7±0.25 <sup>b</sup>	1.0±0.20 <sup>b</sup>	1.1±0.30 <sup>b</sup>

Overall mean values with different superscript alphabets along the same row are significantly different ( $P < 0.05$ ).

**Motor strength (grip time)**

The motor strength values of 1.5 to 2.0 min recorded in groups 4, 5 and 6 rats was significantly ( $P > 0.05$ ) lower than the values of 2.3 to 2.6 min obtained in control and in group 1, 2 and 3 rats (Table 4).

**Table 4: Motor strength (grip time) of male Wistar rats in response to different doses of extracts of *Isoberlinia doka* (for each group n = 5).**

Days	Dose (mg/kg body weight)						
	Control	200	600	1000	1400	1800	2200
1	3.2±0.61	3.3±0.20	2.1±0.50	2.1±0.10	2.4±0.50	1.1±0.01	2.0±0.06
7	1.5±0.10	1.5±0.01	2.0±0.10	1.0±0.01	1.3±0.02	1.5±0.01	1.0±0.50
14	2.2±0.10	3.5±0.20	3.0±0.50	1.0±0.01	1.1±0.02	2.1±0.06	2.0±0.10
21	2.2±0.10	2.1±0.10	3.3±0.15	1.3±0.05	2.5±0.05	1.1±0.02	1.2±0.03
Mean ± SEM	2.3±0.50 <sup>a</sup>	2.6±0.51 <sup>a</sup>	2.6±0.70 <sup>a</sup>	1.4±0.01 <sup>b</sup>	1.8±0.10 <sup>b</sup>	1.5±0.22 <sup>b</sup>	1.6±0.10 <sup>b</sup>

Overall mean values with different superscript alphabets along the same row are significantly different ( $P < 0.05$ ).

In general, there was no significant ( $P > 0.05$ ) different between the results obtained in group 1 and 2 rats and that of the control.

## DISCUSSION

The major task of righting reflex is to maintain the normal standing position and keep an animal head upright. This centre is integrated in the nuclei of the midbrain. Impairment of the righting reflex centre may result to the inability of an animal to righting itself when tilted [11]. The impaired righting reflex observed in rats administered 1400-2200mg of the extract demonstrated that the extract had the ability to damage neuromuscular coordination and sensorimotor reflex, which are the major way responsible for righting reflex coordination. The result of the present finding suggested that the extract may have interfered with the mechanism of neuromuscular transmission of signals and sensorimotor conduction, or it block the righting centre located in the midbrain. Similar observations were made in individuals exposed to agents that perpetuated oxidative damage of the nervous system [11, 12]. The present result further suggests that the administration of higher doses of the extract may have induced free radical generations, which have been implicated in the mechanism of oxidative damage [11, 13]. The result obtained in group 1, 2 and 3 rats compared to control group showed that the extract at 600-1000mg did not have any significant effect on righting reflex of the rats.

The fact that the postural reflex of the rats administered extract of I.D at a dose of 1000-2200mg were between 0-8-1.4 rating showed that the postural reflex was significantly ( $P < 0.05$ ) impaired compared to the control and rats that were given lower doses which had postural reflex of 0-0.4 rating. The result suggested that the extract at a high dose may damage posture regulating pathways. This regulatory pathway is shown to involve the spinal cord, the brain stem and the cerebral cortex [11]. Similar damaging effects on postural reflex were observed in individuals exposed to substances that damage the brain, particularly the cerebellum and cerebral cortex [11, 14]. The obtained result suggested that the extract may have caused the degeneration of motorneuron. The result showed that lower doses between 200-600mg had no effect on the postural reflex of the rats.

The decreased in tactile-placing response observed in groups 5 and 6 rat showed that the extract has impaired forelimb tactile-placing responses, probably by blocking sensorimotor response in the motor cortex and corticospinal tract in the brain which are responsible for limb-placing response [10, 11, 15]. The corticospinal is responsible for skilled movement [11]. The result therefore suggests that ingestion of the extract at higher doses may impair some particular skill performance in individuals.

The significant decrease in forepaw grip time (motor strength) in group 4, 5 and 6 rats suggested a deficit in forepaw motor strength. The result demonstrated that extract of I.D at a dose of 1000 mg and above may reduce the viability of the peripheral nervous system to properly conduct impulses to recap for organs or the inability of the muscles and receptor organs to receive nervous impulse. Studies have shown that agents that will damage microtubule synthesis and functions will result to a decrease in the viability of peripheral nervous system, consequently, a reduction in the ability to grip objects [11, 15]. The result of the present study may suggest that the extract has damage microtubule synthesis and functions. Further investigation is highly required. Ganong [11] has shown that deficits in forepaw grip time are consistent with loss of control of the distal musculature of the limbs, which is responsible for fine skilled movement.

In general the result of the present study, for the first time, showed that higher doses of 1000mg/kg body weight of crude extract of I.D inhibited neuromuscular and sensorimotor pathways, which has resulted to lost of postural stability, skill performance, motor strength and placing response. The lack of information in the available literature on the effect of extract of I.D on behavioural activities did not warrant any comparison of the present result with other studies.

The proximate mechanism of action of the extract on behaviour and the active increment were not investigated, the presence of several secondary metabolites in the extract like, alkaloid, flavonoids, glycoside, tannin and volatile oils may have been responsible for the impairment of behavioural activities. Such secondary metabolites, especially at higher dose have been implicated in the mechanism of free radical generation which has been reported to damage nerve cells [16, 17, 18, 19].

Although folk medicine has reported the use of I.D in the treatment of ailments like hepatitis, nausea, diabetic, infertility etc. it should be used with great caution.

## CONCLUSION

In conclusion, the present study showed that higher doses of I.D above 600mg/kg body weight inhibit neuromuscular and sensorimotor pathways, which have resulted to the impairment of postural stability, skill performance, motor strength, placing and righting reflex responses.

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**REFERENCES**

- [1] O. I. Faleyimu, O. Akinyemi, Y. M. Idris, *Inter. J. Biomed. Hlth. Sci.*, **2010**, 6 (1), 75-81.
- [2] World Bank; Medicinal plants: Local Heritage with global importance. [www.worldbank.org](http://www.worldbank.org). **2001**, Accessed 20/6/2011
- [3] A. Sofowora; Medical plants and traditional medicine in Africa. John Wiley and Sons, New York, 251, **1982**.
- [4] I. O. Lawal, A. B. I. Igboanugo, B. Osikarbor, O. P. Duyilemi, A. A. Adesoga, T. I. Borokini, B. A. Adeyanju, *J. Clin. Med. Res.*, **2010**, 3(4), 061-066,
- [5] D. A. Edwards, B. Walter, P. Liang, *Physiol. Behav.*, **1996**, 60, (5), 1347-1354
- [6] O. I. Faleyimu, O. Akinyemi, *Egyptian J. Biol.*, **2010**, 12, 103-107.
- [7] D. Kubmarawa, G. A. Ajoku, N. M. Enwerem, D. A. Okorie, *Afri. J. Biotechnol.*, **2010**, 6 (14), 1690-1696.
- [8] J. B. Harbone, Phytochemical methods. London, Chapman and Hall Ltd., **1973**, 49-188.
- [9] CCAC; Canadian Council on Animal Care Guide. Vol.1 (2<sup>nd</sup> Ed)., **1993**.
- [10] M. B. Abou-Donia, I. B. Goldstein, K.H. Jones, A. A. Abdel-Rahaman, T. Damodaran, A. M. Dechkovskaia, *Toxicol Sci.*, **2001**, 205-314.
- [11] W. F. Ganong; Review of Medical Physiology 22<sup>nd</sup> edition, McGraw-Hill, London, **2005**
- [12] S. K. Powers, M. J. Jackson, *Physiol. Rev.*, **2008**, 88, 1243-1276.
- [13] K.B. Alok, M. Amritlal, C. J. Dipanjan, C. Sajah, *Mol. Cellular Biochem.*, **2003**, 253, 307-312.
- [14] E. R. Kandel, J. H. Schwaz, In: T. M. Jessell (Ed.), Principle of Neural Science, 4<sup>th</sup> edition, McGraw-HILL, **2000**.
- [15] S. P. Hicks, C. J. D'Amato, *Amer. J. Anat.*, **1975**, 143, 1-42.
- [16] G. E. Trease, W. O. Evans; Pharmacology, 11<sup>th</sup> Edition Belliere Tindal, Lond, Macmillan Publishers, **1989**.
- [17] P. O. Omtayo, O. A. Omoyemi, *Bull. Biol. Sci.*, **2009**, 7 (1), 1-6.
- [18] A. A. Adeiza, N. S. Minka, *Arch. Appl. Sci. Res.*, **2010**, 2 (5): 217-225.
- [19] A. A. Adeiza, N. S. Minka, *Annals of Biol. Res.*, **2011**, 2 (1): 107-113.