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Short Term Toxicological Evaluation of the *Pentaclethra macrophylla* (Benth) Essential Oil (Oil Bean Seed) on Organs of Normal Male Albino Rats

A. M. Yusuf-Babatunde^{1*}, L. S. Kasim² and O. T. Osuntokun³

¹Department of Pharmaceutical Technology, Ogun State College of Health Technology, Ilese-Ijebu, Nigeria.

²Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Olabisi Onabanjo University, Ogun State, Nigeria.

³Department of Microbiology, Faculty of Science, Adekunle Ajasin University, P.M.B 001, Akungba Akoko, Ondo State, Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. Author AMYB designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author LSK managed the analyses of the study. Author OTO managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

The purpose of this research is to evaluate the short term toxicological assessment of the *Pentaclethra macrophylla* (Benth) oil (oil bean seed) on organs of normal male albino rats. Fresh pods of *Pentaclethra macrophylla* (Benth) were collected directly from the plant tree in July 2009 at the site opposite the Bells University of Technology, Ota, Ogun State. Twenty four (24) healthy male albino rats purchased from Animal house of Department of Veterinary Pathology, University of Ibadan, and weighing between 92.0 – 105.4 g were used for the experiment. The effect of the oil extract was investigated by oral administration of the oil into different groups of rats using oral cannular (Group A-D). All the administration was carried out as single dose continually for 8 weeks. The toxicity effects of the extracted oil from *Pentaclethra macrophylla* (Benth) was investigated on

*Corresponding author: E-mail: yusufademol4u@yahoo.com, ogbonronka@gmail.com;

five organs of rats (liver, brains, kidney, heart and testis). The animals were divided into four groups (A, B, C, and D) fed with a normal diet and three different levels of concentrations of the oil. With the oral cannular techniques, the first group (A) rats were fed with 0.2 ml/day of oil with a normal diet, second group (B) with 0.4 ml/day with a normal diet, the third group (C) fed with 0.6 ml/day oil with normal diet while fourth group D were treated with normal diet only, serving as the control group for the study. The results of the histological examination indicated that as the concentrations of the oil ingested increases, some of the tissues of the organs were affected. Therefore, the study suggested that *Pentaclethra macrophylla* (Benth) oil was toxic to the organs of the rats. Therefore, the consumption of the seeds by the human can constitute health hazards and risks.

Keywords: Toxicity; Pentacletra macrophylla; organs; histology.

1. INTRODUCTION

Toxicity of fixed oil is the degree to which oil is poisonous to living cells. The fixed oils and fats are mixtures of three bodies, Olein (Liquid), Palmitin (Semisolid) and Stearin (Solid) or close relations of these and also a usually small amount of other bodies. Olein, palmitin and stearin are compounds of glyceryl with radicles of the various fatty acids with alkalies, they form soaps and glycerine. Castile soap, for example, is made by the action of sodium hydroxide on olive oil, which is nearly pure Olein. A few of the fats and oils are of animal origin but the majority are of vegetable origin. Some fats are essential for proper brain function. Two lipids important to the brain are the n-6 and n-3 fatty acids. Low levels of n-3 fatty acids in a diet can cause visual problems especially by affecting the retina. Studies in animals have shown that diets without n-3 fatty acids cause learning, motivation and motor problems and may affect systems that use the neurotransmitters dopamine and serotonin in the frontal cortex. The n-6 fatty acids are also important in proper brain function because they affect neurotransmitter release and they also influence the ability of neurons to use glucose [1].

Fats and fixed oils are used as essential foods and some as solvents in the intramuscular injection (e.g. sesame oil), other have medicinal action e.g. castor oil (as a cathartic) and liver oil (as an emollient), [2,3].

Penthacletra macrophylla (Benth) is also known as the Africa oil bean tree, a leguminous tree (family Leguminosae, subfamily Mimosoideae) has been cultivated in Nigeria for many years and in other West African Countries where its seed is relished as food. The local names include "congo acacia" in congo, "Duala Kombola" in Cameroon and "Ugba", "Ukpala" and "Apara" in South Eastern and western Nigeria. Fermented *Penthaclethra macrophylla* (Benth) seed is a very important delicacy in the life of Nigerians. Its use as food among Eastern populace of Nigeria and as a delicacy across the different tribes in Nigeria. The oil bean seeds are inedible when raw and bitter until the final stage of fermentation. The fermentation improves flavour and taste and has been described as an economic processing method used in homes to improve nutritional quality [3,4].

The bean essential oilseed is rich in protein and minerals therefore it serves as a cheap source of protein thus increasing the haematological parameters [5]. It has been observed that there was a depression in weight along with negative protein utilization which may also lower haematological parameters in rats fed with various tropical legume seed including oil bean, they attributed those adverse effects to the presence of heat-stable toxicants in the seeds, [6]. Also, some anti-nutritional factors have been identified and assayed in the Pentachletra macrophylla [7]. The present study has therefore been conducted to examine the toxicological effect of the oil of Pentachletra machrophylla Benth on organs of rat (namely; heart, kidney, liver, brain and testis).

2. MATERIALS AND METHODS

2.1 Sources of Seeds

Fresh pods of *Pentaclethra macrophytlla* (Benth) were collected directly from the plant tree in July 2009 at the site opposite the Bells University of Technology, Ota, Ogun State and authenticated at the herbarium of the Forestry Research Institute of Nigeria (FRIN), Ibadan. (Voucher No. FH1108453). *Pentaclethra macrophylla* (Benth) (1.0 kg) semi-dried seeds were ground into smaller pieces and macerated in 3L of n-hexane for 72 hours. It was filtered and the liquid extract concentrated in rotary evaporated at 35°C leaving the concentrated oil. The oil was stored at 4°C and used when required [8].

2.2 Test Experimental Subjects

Twenty four (24) healthy male albino rats purchased from Animal house of Department of Veterinary Pathology, University of Ibadan, and weighing between 92.0 - 105.4 g were used for the experiment. The rats were divided into four groups of six rats per group. All the animals were acclimatized for 7 davs before the commencement of the experiment. Rats were kept in cages under standard conditions and fed with pellets (Livestock Nigeria Plc, Lagos) and water ad libitum.

2.3 Administration of the Extract

The effect of the oil extract was investigated by oral administration of the oil into different groups of rats using oral cannular. The group were labelled and treated as indicated below

- (i) A 0.2 ml daily for 8 weeks + normal diet
- (ii) B 0.4 ml daily for 8 weeks + normal diet
- (iii) C 0.6 ml for 8 weeks + normal diet
- (iv) D (Control test group) treated with a normal diet.

All the administration was carried out as single dose continually for 8 weeks. The animals fasted for 24 hours after 8 weeks pre-treatment period.

All the rats were anaesthetized by keeping them in an enclosed container containing chloroform. Blood was withdrawn by cardiac puncture into EDTA sample bottles and Lithium heparin bottles for biochemical analysis. The animals were open up (dissected) and the heart, liver, kidney, testis and the brain were removed and put in bottles that contain 4% formaldehyde in phosphate buffer and histology test of the tissues of the organs were carried out at the department of veterinary medicines, University of Ibadan, Ibadan, Nigeria [9].

3. RESULTS AND DISCUSSION

The purpose of this research is to evaluate the short term toxicological assessment of the *Pentaclethra macrophylla* (Benth) oil (oil bean seed) on organs of normal male albinorats.

From the histological examination of five organs of rats, (brain, liver, kidney, heart and testis), it was observed at 0.2 ml/day treatment, there was no harmful damage in the cells of brains, kidney,heart and testis. There was portal congestion, cellular infiltration, an abnormal increase in the number of cells at bile duct in the liver. At 0.4 ml/day treatment (group B), there were no harmful damages in the cells of brains and testis but there was portal congestion blood), cellular (blocked with infiltration (penetration of cells) in the liver of rats, haemorrhage (flow of blood from a ruptured blood vessel) in the parenchyma of the heart of one rat. Three rats had vascular congestion (blocked with blood) at the cortex of the kidney [10].

Osuntokun [11], reported that the no alteration observed in the liver and kidney, the observable features are dose and time-dependent. The presence of nephritic necrosis is not unexpected

 Table 1. Effect of Pentaclethra macrophylla (Benth) essential oil on the organs of male albino rats with 0.2 ml/day treatment

Sample	Brain	Liver	Kidney	Heart	Testis
A1	No visible	Mild portal cellular	No visible	No visible	No visible
	lesions seen	infiltration, bile duct hyper-lasia	lesions seen	lesions seen	lesions seen
A2	No visible	No visible lesions	No visible	No visible	No visible
	lesions seen	seen	lesions seen	lesions seen	lesions seen
A3	No visible	Multifocal, portal	No visible	No visible	No visible
	lesions seen	cellular infiltration	lesions seen	lesions seen	lesions seen
A4	No visible	Severe central venous	No visible	No visible	No visible
	lesions seen	and portal congestion	lesions seen	lesions seen	lesions seen
A5	No visible	No visible lesions	No visible	No visible	No visible
	lesions seen	seen	lesions seen	lesions seen	lesions seen
A6	No visible	No visible lesions	No visible	No visible	No visible
	lesions seen	seen	lesions seen	lesions seen	lesions seen

Sample	Brain	Liver	Kidney	Heart	Testis
B1	No visible	Multifocal of	No visible	No visible	No visible
	lesions seen	mononuclear cellular infiltration	lesions seen	lesions seen	lesions seen
B2	No visible lesions seen	No visible lesions seen	Vascular congestion at the cortex	No visible lesions seen	No visible lesions seen
B3	No visible lesions seen	Portal congestion and cellular infiltration by mononuclear cells	Vascular congestion at the cortex	Multiple foci of haemorrhages in the parenchyma	No visible lesions seen
B4	No visible lesions seen	Portal congestion and cellular infiltration by mononuclear cells	Vascular congestion at the cortex	No visible lesions seen	No visible lesions seen
B5	No visible lesions seen	No visible lesions seen		No visible lesions seen	No visible lesions seen
B6	No visible lesions seen	Multifocal and <i>periportal</i> cellular infiltration by mononuclear cells	No visible lesions seen	No visible lesions seen	No visible lesions seen

Table 2. Effect of Pentaclethra macrophylla (Benth) essential oil on organs of male albino rats with 0.4 ml/day treatment

Table 3. Effect of Pentaclethra macrophylla (Benth) essential oil on organs of male albino rats with 0.6 ml/day treatment

Sample	Brain	Liver	Kidney	Heart	Testis
C1	No visible lesions seen	Portal congestion and cellular infiltration by mononuclear cells	No visible lesions seen	No visible lesions seen	No visible lesions seen
C2	No visible lesions seen	Multifocal portal cellular infiltration	Protein casts in the tubular lumen	No visible lesions seen	Severe interstitial <i>oedema,</i> severe germinal erosions
C3	No visible lesions seen	No visible lesions seen	Cortical haemorrhages and interstitial cellular infiltration	Marked haemorrhages	<i>Necrotic</i> debris in the seminiferous tubular lumen
C4	No visible lesions seen	Severe congestion (central venous) and portal cellular infiltration	Cortical haemorrhage	No visible lesions seen	No visible lesions
C5	No visible lesions seen	No visible lesions seen	No visible lesions seen	No visible lesions seen	No visible lesions seen
C6	No visible lesions seen	Cellular infiltration by mononuclear cells (diffuse)	No visible lesions seen	Marked congestion and haemorrhages	Severe interstitial oedema

Since excretion is mainly carried out in the kidney; the observation found in the kidney of the extracts treated rats is following the finding in the

study that reported that no tubular necrosis occurred in rats fed with anti-nutritive factors containing feeds.

At 0.6ml/day (group C), there was no harmful damage in the cells of the brain, there was severe interstitial oedema (accumulation of fluid) in the testis of two rats, necrotic debris (injury which results in the premature death of cells) was observed in the seminiferous tubular lumen in the testis of one rat. In the heart of two rats were observed marked haemorrhage (flow of blood from a ruptured blood vessel). In the kidney of two rats were observed cortical haemorrhage and protein casts in the tubular lumen of one rat. In the liver of four rats were observed portal cellular infiltration (penetration of cells in an area) [3].

Sample	Brain	Liver	Kidney	Heart	Testis
D1	No visible	Marked central venous	No visible	No visible	No visible
	lesions seen	and portal congestion with multifocal cellular infiltration	lesions seen	lesions seen	lesions seen
D2	No visible	Cellular infiltration by	No visible	No visible	No visible
	lesions seen	mononuclear cells	lesions seen	lesions seen	lesions seen
D3	No visible	No visible lesions seen	No visible	No visible	No visible
	lesions seen		lesions seen	lesions seen	lesions seen
D4	No visible	Severe congestion	No visible	No visible	No visible
	lesions seen	(central venous) and	lesions seen	lesions seen	lesions seen
		portal cellular infiltration			
D5	No visible	Mild portal congestion	No visible	No visible	No visible
	lesions seen	and cellular infiltration	lesions seen	lesions seen	lesions seen
C6	No visible	Multifocal cellular	No visible	No visible	No visible
	lesions seen	infiltration	lesions seen	lesions seen	lesions seen



Fig. 1a. Sample test (C) of brain

Fig. 1b. Control test (D) of brain



Fig. 2a. Sample test (C) of liver



Fig. 2b. Control test (D) of liver

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Fig. 3a. Sample test (C) of kidney

Fig. 3b. Control test (D) of kidney



Fig. 4a. Sample test (C) of heart

Fig. 4b. Control test (D) of heart



Fig. 5a. Sample test (C) of testes



Fig. 5b. Control test (D) of testes



Fig. 6a. Sample test (B) of brain

Fig. 6b. Control test (D) of brain

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Fig. 7a. Sample test (B) of liver

Fig. 7b. Control test (D) of liver



Fig. 8a. Sample test (B) of kidney

Fig. 8b. Control test (D) of kidney

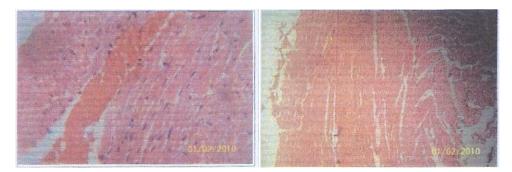


Fig. 9a. Sample test (B) of heart

Fig. 9b. Control test (D) of heart



Fig. 10a. Sample test (B) of testes

Fig. 10b. Control test (D) of testes

Figs. 1- 10. Histology test results of the *Pentaclethra macrophylla* (Benth) oil on the tissues of rats

In the control animals (group D), it was observed that there was no harmful damage seen in the brains, kidneys, hearts and testis of all rats as was the case in group A. There was portal congestion, cellular infiltration in the livers of rats as also observed in group A. From the observations of the test it is shown that as the concentration of the ingested oil of *Pentraclethra macorphylla* increases there was the increase in its harmful effects in organs of rats.

4. CONCLUSION

This study reports the noticeable toxic effect of the oil of *Pentaclethra macrophylla* on organs of rats. The results showed that the oil at higher concentrations is toxic to the organs of rats. It, therefore, suggests that caution should be observed in the administration of the *Pentaclethra macrophylla* oil in human being.

CONSENT

It is not applicable.

ETHICAL APPROVAL

As per international standard or university standard written ethical permission has been collected and preserved by the author(s).

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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