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Toxicity study of ethanol root extract of *Terminalia macroptera* Guill. & Perr. (*Combretaceae*) and assessment of some heavy metals

Y. Yakubu[1]*, O. A. Adoum[2], A. M. Wudil[3] and Z. Ladan[1]

[1]Department of Chemistry, Faculty of Science, Kaduna State University, Kaduna State, Nigeria.
[2]Department of Pure and Industrial Chemistry, Faculty of Science, Bayero University, Kano, Kano State, Nigeria.
[3]Department of Biochemistry, Faculty of Science, Bayero University, Kano, Kano State, Nigeria.

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The extract of *Terminalia macroptera* Guill. & Perr. has been used in the management of pile and other ailments across West Africa. This has necessitated the evaluation of the toxicity of this plant in Sub-chronic and acute administration as well as heavy metal analysis. The root of *T. macroptera* was extracted with ethanol and analyzed for the presence of phytochemicals. The powdered root was digested and analyzed for presence of Cd, Cr, Pb, Ni and Zn using spectrophotometric technique. The sub-acute toxicity on some liver parameters was evaluated following the administration at doses of 1.5, 1.0 and 0.5 g/kg respectively. The phytochemical screening of the extract revealed the presence of alkaloids, flavonoids, steroids, glycoside, resins and saponin. No death of a single animal was recorded after the acute toxicity study. The effect of sub-chronically administered extract was P< 0.05 of ALT, AST and ALP at different doses when compared to the control. The quantity of metal detected was; Cd (0.0830 ppm), Cr (0.3516 ppm), Pb (0.8809 ppm), Ni (0.6438 ppm) and Zn (0.7340 ppm), which are all within the normal range (1 ppm) as recommended by the Federal Ministry of Environment, Nigeria.

Key words: Acute toxicity Heavy metals phytochemicals *Terminalia macroptera* Guill. & Perr., Sub-chronic toxicity.

INTRODUCTION

Terminalia macroptera Guill. & Perr. (Combretaceae) is a tree, up to 20 m high, which occurs widely in West Africa. In Mali *T. macroptera* is used against a variety of ailments, and more than 30 different indications have been mentioned by the traditional healers in ethno-pharmacological studies. The stem bark and leaves are most commonly used against sores and wounds, pain, cough, tuberculosis and hepatitis (Pham et al., 2011). The roots are used against hepatitis, gonorrhea and various infectious diseases, including *Helicobacter pylori* -associated diseases (Silva et al., 2012, 2000). Root bark, stem bark and leaves are used in traditional medicine in Nigeria against several ailments.

Heavy metals with adverse health effects in human metabolism (including lead and cadmium) present obvious concerns due to their persistence in the

*Corresponding author. E-mail: bbmobly@yahoo.com

Table 1. Summary of phytochemical screening test.

Plant fraction	Ethanol
Saponins	+
Alkaloids	+
Tannins	+
Glycocides	-
Flavonoids	+
Resins	+
Steroids	+

+, Present.

environment and documented potential for serious health consequences (ATSDR, 2004, 2007a, b, 2008a, b, 2011).

Acute heavy metal intoxications may damage central nervous function, the cardiovascular and gastrointestinal (GI) systems, lungs, kidneys, liver, endocrine glands, and bones (Jang and Hoffman, 2011; Adal and Wiener, 2013). Chronic heavy metal exposure has been implicated in several degenerative diseases of these same systems and may increase the risk of some cancers (Galanis et al., 2009; Wu et al., 2012). Heavy metals are ubiquitous in the environment (Pohl et al., 2011). Humans risk overexposure from environmental concentrations that occur naturally (eg, arsenic-rich mineral deposits) or human activities (eg, lead or mercury release as a result of industrial pollution) (Orloff et al., 2009; Hutton, 1986). It is not possible to completely avoid exposure to toxic metals (Singh et al., 2011). Even people who are not occupationally exposed carry certain metals in their body as a result of exposure from other sources, such as food, beverages, or air (Washam, 2011; Satarug et al., 2010).

In this study, we have carried out the sub-acute as well as acute toxicity study and assessment of some heavy metals (Pb, Cr, Cd, Zn and Ni) using spectrophotometric technique to check it's level in the root of *T. macropetera* and this will help ascertain total safeness because of its wide consumption by people of that community for treatment of pile and other diseases.

MATERIALS AND METHODS

Collection of the plant

The roots of the plant were collected from Ede Alaba bush in Idah local government area of Kogi state, Nigeria on the 20th February, 2011. It was taxonomically identified by Mallam Mohammed Musa of the herbarium unit Ahmadu Bello University Zaria, Nigeria as *T. Macroptera* (Guill. & Perr.).

Extraction and isolation

The roots were dried in shade, pulverized to powder using pestle and mortar in the laboratory. Approximately 500 g was percolated for 7 days then filtered using whatman filter paper. The filtrate was concentrated and the extract was kept in the refrigerator (4°C) until needed for further use.

Phytochemical screening

The phytochemical analysis of the crude extract was carried out according to standard methods (Harborne, 1998; Soforawa, 1993) (Table 1).

Sub-acute toxicity study

Sub-acute toxicity study was carried out using the ethanol root extract of the plant. Sixteen Wister albino rats were used for this experiment and were grouped into four. Group A was the control group while group B, C and D were orally administered 0.5, 1.0 and 1.5 g/kg ethanol root extract respectively. Blood samples were collected after three weeks of administration by scarification according to WHO standards. Serum sample collected were used for liver function test (Table 2).

Acute toxicity study

The acute toxicity study was conducted in two phases using a total of twelve rats. In the first phase, nine rats were divided into 3 groups of 3 rats each. Groups 1, 2 and 3 animals were given 10,100 and 1000 mg/kg body weight (b.w.) of the extract, respectively, to possibly establish the range of doses producing any toxic effect. Each rat was given a single dose.

In the second phase, further specific doses (1600, 2900 and 5000 mg/kg b.w.) of the extract were administered to three rats (one rat per dose) to further determine the correct LD50 value. The extract was dissolved in normal saline water and given via intraperitoneal route. All animals were observed frequently on the day of treatment and surviving animals were monitored daily for 2 weeks for signs of acute toxicity (Lorke, 1983).

Heavy metal analysis

Finally the powder (1.0 g) was digested (Dokiya et al., 1975) and analyzed for the presence of Cd, Cr, Pb, Ni and Zn due to their potential health risk.

RESULTS AND DICUSSION

The use of medicinal plants in the treatment of ailments in both the developed and developing countries is on the increase. The therapeutic values of most of these herbs are indisputable but their toxicities sometimes limit their clinical uses. Thus the toxicity of these herbs must always be considered especially as the doses and dosing regimens of their preparations are not usually determined (Chan, 1997). The effect of the sub-chronic administration of the extract on some liver function parameters of rats was determined and the result presented in Table 3. The extract induced significant increase in the levels of ALT, AST and ALP. Serum level of ALT, AST and ALP were dose related but were not significantly ($P< 0.05$) different. Result of the liver function indices were expressed as mean ±SEM. The acute lethal effect (Table 4) of ethanol root extract of *T. macroptera*

Table 2. Results of sub-acute test.

Group	ALT(U/L)	AST(U/L)	ALP(U/L)	DOSE(mg/kg)
A1	108	159	68	
A2	69	169	15	
A3	55	189	19	Control group
A4	73	162	22	
B1	70	202	20	
B2	113	205	84	
B3	65	175	16	1.5 mg/kg
B4	70	189	16	
C1	61	223	21	
C2	130	192	76	
C3	62	174	21	1.0 mg/kg
C4	79	185	16	
D1	70	180	28	
D2	133	188	72	
D3	83	292	31	0.5 mg/kg
D4	82	199	21	

Table 3. Analyzed sub-acute toxicity test.

Group	ALT	AST	ALP
A	76 ± 22.64	169 ± 13.34	31 ± 24.83
B	79.5 ± 22.45	192.8 ± 13.75	34 ± 33.39
C	83 ± 32.40	193.5 ± 21.0	33.5 ± 28.43
D	92 ± 27.96	214.8 ± 52.08	36.8 ± 23.89

Mean ± Standard Deviation.

Table 4. Acute lethal result of *T. maroptera* Guill. & Perr. administered intraperitoneally.

Experiment	Dose (mg/kgbw)	No of rats after 24 h	Treated rats after 24 h
Phase-1	10	0/3	0/3
	1,00	0/3	0/3
	1,000	0/3	0/3
Phase 2	1,600	0/1	0/1
	2,900	0/1	0/1
	5,000	0/1	0/1

induced intraperitoneally showed that no animal died after 24 hours. The major sign of toxicity noticed was general weakness, and loss of appetite. The sign became increasingly pronounced as the dose increased towards 5000 mg/kg. The signs were not noticed in 10 and 100 mg/kg doses respectively. The LD50, being greater than 5000 mg/kg b.w., is thought to be safe as suggested by Lorke (1983). Again, the absence of death among rats in all the dose groups throughout the two weeks of the experiment seems to support this claim. Phytochemical analysis of the extract reveals the presence of alkaloid, flavonoid, tannins, steroid, resin and saponin. Just as reported by Nongonierma (1990), that; Flavonoids, have been identified from different parts of *T. macroptera*. The

Table 5. Result of amount of heavy metals present.

Test	Amount(ppm)
Cd	0.0830
Cr	0.3516
Pb	0.8809
Ni	0.6438
Zn	0.7340

presence of these phytochemicals in the crude extract lends credence of the use of the plant for medicinal purposes. The use of some plants for medicinal purpose, in traditional treatment of diseases is due to the presence of flavonoid and saponins (Zwadyk, 1992; Othira et al., 2009) hence the use of *T. macroptera* Guill. & Perr. for treatment of hepatitis, ulcer, hemorrhage, diarrhea and several other ailment by traditional healers is not surprising. Tannins in some medicinal plants have been found to be responsible for the antiviral and antibacterial activities exhibited by such plant (De-Ruiz et al., 2001; Elegani et al., 2002) The result of the heavy metal analysis revealed that the amount Cd (0.0830), Cr(0.3516), Pb(0.8809), Ni(0.6438) and Zn(0.7340) were all within the limit of safety set by federal ministry of environment of Nigeria (Table 5). In conclusion, the plant is safe for consumption if plant source is harvested from areas not prone to industrial waste or areas exposed to hazard that can cause heavy metal contamination. Further work is recommended on this plant to determine its anti-pile activity claimed by some locals in Nigeria.

Conflict of Interest

The authors have not declared any conflict of interest.

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