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Investigation of Acute Toxicity Level of Gongronema Latifolium Leaf Extract on White Albino Rat

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Abstract

The aim of this study is to investigate the level of toxicity in *Gongronema latifolium* leaf extract on male swiss albino rats. Lorke's method of toxicity was used in this research study. The experimental animals were administered with different doses of the extract. The rats were monitored closely for behavioural changes, difficulty in breathing and death for the period of two weeks. At the end of the two weeks study, all the animals used for the experiment were sacrificed and the internal organ-body weight ratio (OBR) were determined and compared with the values from the control group. The LD50 was found to be >5000 mg/kg body weight. There was no significant weight decrease ($P>0.05$) among dose groups of up to 1000mg/kg body weight. Liver congestion was observed with 100mg/kg body weight dose group. The OBR mean values for kidney, liver, and heart were not significantly ($P>0.05$) different from that of the control group. The extract therefore is safe for human consumption.

Keywords: acute toxicity; *gongronema latifolium*; albino rats; traditional medicine

Introduction

Traditional medicine has a long history. It is the sum total of the knowledge, skill, and practices based on the theories, beliefs, and experiences indigenous to different cultures, whether explicable or not, used in the maintenance of health as well as in the prevention, diagnosis, improvement or treatment of physical and mental illness [1].

An estimated 400 million inhabitant of the world, that is about 80% of world's population, are thought to rely chiefly on traditional medicine, which is largely of plant origin, for their primary health care needs [2]. However, it is widely believed that this valuable medicinal resources in plants are largely untapped because of inadequate scientific technical and commercial infrastructure in developing countries [3]. In recent years, there is a growing interest therapy. Data on scientific screening of plant extracts, whether crude or purified, appeared to be accumulating gradually but steadily. Literatures on antidiarrheal, antimalarial and antitrypanosomally activities of plant-based products support this claim [4-7]. The major contributory factors to this growing interest include: rising costs of orthodox medicines, low therapeutic index of synthetic compounds and the growing incidence of drug resistance [8,9] among the pathogens especially in developing countries with very weak economic indices. It is thought that the use of plant-derived active principles will offer people access

to safe and effective products for prevention and treatment of diseases through self-medication [10].

Gongronema latifolium (Asciadiaceae) utazi in igbo and bushbuck in English is an edible rain forest plant, native to South Eastern part of Nigeria. It has been widely used in folk medicine as spices and vegetable and for maintaining blood glucose levels [11].

The aim of this study is to evaluate the toxic potential of this plant with the view to endorse or refute the safety use of its crude extract in traditional medicine.

Materials and Methods

Fresh and healthy leaves of the medicinal plants of *Gongronema latifolium* were collected from the plants naturally grown in Umuojogwo Village Umuchu, Aguata Local Government area of Anambra State, Nigeria. The specimens were identified at the Department of Pharmacognosy and Traditional Medicine of Nnamdi Azikiwe University Awka, Agulu Campus, Anambra State Nigeria.

Preparation of Crude Extract

The *Gongronema latifolium* leaf were thoroughly washed in distilled water and a known quantity (400g) were dried at room temperature for 7 days. Four hundred (400) grams of the pulverized plant sample was mascerated in 1000ml (1 liter) of 95% methanol over a period of 48 hours with intermittent shaking. The mixture was sieved using muslin cloth. It was further filtered using No.1 whatman filter paper. The

filtrate was concentrated using rotary evaporation at reduced temperature and pressure (100C). It was further concentrated using water bath at 500c. The crude extract was stored in refrigerator for use.

Experimental Animals

Fifteen white male albino rats (Wister stock) were obtaining from the Department of Pharmacology and Toxicology, Nnamdi Azikiwe University Agulu Campus, Awka Anambra State Nigeria. The animals were fed on diet specially prepared from Chick Grower's mash (Pfizer Company, Nigeria) and was given water *ad libitum* throughout the study period. Before the commencement of the experiment, the weight of the animals ranged from 130g to 200g.

Experimental Design for Acute Toxicity Study

The acute toxicity study was done using the method employed by [12]. A total of 16 male rats was used in this study. The study was done in two phases. In phase 1, nine rats were grouped into 3 rats per group. Group one received 10mg/kg, group two received 100mg/kg while group three group received 1000mg/kg, this is done to possibly establish the range of doses producing any toxic effect. Each rat was given a single dose after at least 5 days of adaptation. In addition, a fourth group of three rats was set up as control group and animals in the group were not given the extract. In the second phase, further specific doses (1600, 2900 and 5000mg/kg b.w.) of the extract was administered to three rats (one rat per dose) to further determine the correct LD50 value. The extract was dissolved in phosphate buffered saline (PBS) solution 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. Recovery and weight gain were seen as indication of having survived the acute toxicity. At the end of 14 days, all surviving rats were

sacrificed and then autopsied at Springboard pathology laboratory Awka, Anambra State Nigeria and the internal organs were examined macroscopically for pathological changes compared to the control group. The weight of these organs was also taken and the mean organ-body weight ratios calculated and compared with those of the control group.

Statistical Analysis

The statistical analyses were carried out using statistical package for social sciences (SPSS-Computer Package). Percentage organ-body weight ratios and rats' body weights were expressed as mean $\pm$ SD. Values in all groups were compared using the analysis of variance (ANOVA). For all analyses the level of statistical significance was fixed at $p < 0.05$ [13].

Results

The acute toxicity study of *Gongronema latifolium* leaf extract on rats (Table1) shows that no animal died within 24 hours after administering extract and the LD50 was greater than 5000mg/kg b.w. Again, no death was recorded among all the dose groups throughout the two weeks of the experimental period. Observation: The LD50 > 5000 mg/kg b.w. Furthermore, a dose-dependent weight loss occurred but the weight variations noticed among the extract treated groups (Table 2) were not found to be significant ($P > 0.05$) when compared with the control group. Table 3 gives the gross pathological features of some internal organs. Congested liver was seen in all rats treated with the extracts. This observation appears to be in harmony with the OBR values given in Table 4.

Table 1: Acute Lethal Effect of *Gongronema latifolium* leaf Extract Administered Intraperitoneally (IP) to White Albino Rats.

Experiment	Dose (mg/kg b.w.)	No Dead Rats After 24 Hours	Treated Rats After 24 Hours
Phase 1*	10	0/3	0/3*
	100	0/3	0/3
	1000	0/3	0/3
Control	0	0/3	0/3
Phase 2	1600	0/1	0/1
	2900	0/1	0/1
	5000	0/1	0/1

Table 2: Effects of Intraperitoneal Administration of *Gongronema latifolium* Leaf Extract on the body weight of Rats during Acute Toxicity test.

Experiment	Dose (mg/kg b.w.)	Weight gain (x±SD)
Phase 1	10	42.80±5.05b
	100	38.37±9.87b
	1000	31.63±4.63±4.68b
Control	0	40.85±14.27b
Phase 2	1600	14.50
	2900	5.80
	5000	4.70

Table 3: Post Mortem Result for Acute toxicity of *Gongronema latifolium* Leaf Extract Administered intraperitoneally (IP) to White Albino Rats.

Gross Pathology Observed Dose (mg/kg b.w.)				
Organ	0	10	100	1000
Liver	None	Congested	Congested	Congested
Lungs	None	None	None	None
Kidney	None	None	None	None
Heart	None	None	None	None
Spleen	None	None	None	None

Table 4: Effects of Intraperitoneal (I.P) Treatment with *Gongronema latifolium* Leaf Extract on percent organ-body weight ratios of rats after the acute toxicity test.

Organ-Body Weight Ratio Dose (Mg/Kg B.W)							
Organ	10	100	1000	1600	2900	500	Control
Kidney	0.3500±0.0255	0.3776±0.701	0.4300±0.0000	0.3600	0.2800	0.29	0.3621±0.0647
Liver	3.5777±0.5736	3.6680±0.7220	4.8000±0.4646	5.0000	3.5000	4.9	4.0500±0.4473
Lungs	0.5200±0.0822	0.777±0.0924	0.9200±0.0525	0.5900	0.7000	0.88	0.8000±0.1339
Spleen	0.4222±0.1006	0.6600±0.2365	0.3740±0.0525	0.3600	0.2600	0.38	0.3050±0.1035
Heart	0.3231±0.0260	0.2760±0.0040	0.28400±0.0060	0.3700	0.2400	Dead	0.3039±0.0321

Discussion

The acute toxicity effect of *Gongronema latifolium* leaf extract on rats (Table 1) shows that no experimental animal died within 2 hours after treatment with the extract. Major signs of toxicity noticed within 24 hours included difficulty in breathing, loss of appetite and general weakness. These signs were not seen in 10mg/kg b.w. dose group but progressed and became increasingly pronounced as the dose increased towards 5000mg/kg b.w. The LD₅₀ being greater than 5000mg/kg b.w., is thought to be safe as suggested by [12]. Again, the absence of death among rats in all the dose groups throughout the two weeks of the experiment seems to support this claim. Furthermore, the dose-dependent weight loss observed, were not found to be statistically significant ($P>0.05$) when compared with the control group (Group 2).

Liver congestion appeared to be the major gross pathology accompanying treatment of rats with

Gongronema latifolium leaf extract (Table 3). This observation is in agreement with the increased OBR values given in Table 4, though the values were not statistically significant ($P>0.05$). When compared with [14], extract from *Gongronema latifolium* appears safer for usage in traditional medicine. It is possible that this variation is due to the quantitative variation of saponin in these plants as the toxic potential of *Gongronema latifolium* was attributed to saponin in it [14]. Saponins are known to have deleterious hemolyzing effect on circulating erythrocytes [15,16] and their presence in crude extract of *Gongronema latifolium* accounted for its low therapeutic index found against ascariasis infection in mice [21]. Again, liver congestion could be attributed, in part, to its role in biotransformation of xenobiotics. It is not clear why the administration of 100mg/kg b. w., and not higher doses, increased the size of the spleen significantly.

Survey by [17] has shown that *Gongronema latifolium* is one of the herbs commonly used to treat

helminthiasis in Eastern Nigeria. However, there are few scientific literatures to support this [18] [19,20]. To what extent ingestion of extracts from this plant will be toxic is yet to be ascertained.

Conclusion

From the results of this study, it is observed that *Gongronema latifolium* leaf extract is safe for use in traditional medicine. Higher dose should however, be avoided and users should not rule out completely the possibility of chronic toxicity developing with the continual use of *Gongronema latifolium* leaf extract.

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