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Short Communication

Effects of Landolphia owariensis leaf extract on the liver function profile and haemoglobin concentration of albino rats.

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The effects of aqueous extract of *Landolphia owariensis* leaves on the biochemical indices of liver function were investigated in Wister strain of albino rats. Preliminary phytochemical analysis of the plant leaves showed the presence of tannins, alkaloids, flavonoids and saponins. Acute toxicity tests of the extract gave an LD₅₀ of 3370 mg/kg. Liver function tests revealed that the serum activities of alanine aminotransferrase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP), as well as the concentrations of total protein and albumin were not significantly (p>0.05) affected by the oral administration of the extract. However, bilirubin and haemoglobin concentrations decreased significantly (p<0.05) on administration of the extract. The results suggest that *L. owariensis* leaf extract is not hepatotoxic in rats. The findings are of clinical importance given the various reported therapeutic potentials of the plant.

Key words: Landolphia owariensis, hepatotoxicity, LD₅₀, haemoglobin.

INTRODUCTION

Landolphia owariensis is a common African plant that has been found useful due to its therapeutic potentials (Gill, 1992; Owoyele et al., 2002). It belongs to the family Apocynaceae commonly called vine rubber and known locally by various names in Nigeria: Ibo – *Eso/utu*, Yoruba – *Mba* and *Hausa* – *Ciwa*. It is commonly found in the rain forest region of Nigeria and other parts of African countries. *L. owariensis* is one of the plants whose leaves, bark and roots are used for the treatment of many ailments. The decoction from the leaves is used as purgative and to cure malaria. The aqueous, methanolic and chloroform extracts of *L. owariensis* leaves have antimicrobial activities (Owoyele et al., 2002; Nwaogu et al., 2007). Lewis and Lewis (1977) also reported the use of the extract from the stem bark as vermifuge.

The array of applications of *L. owariensis* is hardly surprising, considering that plants have always been the principal source of medication either in the form of traditional preparation or as a pure active principle (lwu,

1992; Sofowara, 1982). These medicinal importance of plants have been attributed to their phytochemical content. Thus phytochemical analysis of plants is predicated by the need for drug alternatives of plant origin, made imperative by the high cost of synthetic drugs. *L. owariensis* leaves have been reported to contain various secondary plant metabolites of medicinal value including saponins, tannins alkaloids and flavonoids. These secondary plant metabolites extractable by various solvents exhibit varied biochemical and pharmacological actions in animals when ingested (Trease and Evans, 1996). This paper reports the effects of aqueous extract of *L. owariensis* leaves on the biochemical indices of liver function and haemoglobin concentrations of albino Wister rats.

MATERIALS AND METHODS

Plant Material

Fresh leaves of *L. owariensis* were collected from their natural habitat in Ikeduru, Imo State, Nigeria, in the month of September, 2007. The plant was identified by Prof. S. E. Okeke, a Plant taxono-

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Table 1. Phytochemical profile of the aqueous leaf extract of Landolphia owariensis

Phytochemical components	Landolphia owariensis extract		
Alkaloids	+		
Flavonoids	+		
Tannins	++		
Saponins	+		
Cyanogenic glycosides	ND		

Keys: ++ = Highly present, + = present, ND = Not Detected.

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Extract preparation

The fresh leaves of *L. owariensis* were washed and sun-dried for ten days to a constant weight. The dried leaves were ground into powder using a mechanical grinder. One hundred grams (100 g) of the powder was weighed and soaked in 1000 ml of pre-boiled distilled water. This was covered, shaken vigorously every 30 min for 2 h and then allowed to stand for 48 h. The solution was subsequently shaken and filtered using Whatman Number 1 filter paper. Extract was concentrated in vacuum and freeze dried (yield, 6.2%). The dried extract was stored in a descicator protected from light and moisture.

Phytochemical studies

Phytochemical test for the presence of alkaloids, flavonoids, saponins, tannins and cyanogenic glycosides were carried out as described by Harborne (1973) and Trease and Evans (1996).

Acute toxicity tests

The acute oral toxicity study was determined in mice. Eight groups of five mice each were orally administered with different concentrations of 0, 62.5, 125, 250, 500, 1000, 2000 and 4000 mg/kg of *L. owariensis* aqueous extract with the aid of an intubator. The animals were observed continuously for 1 h and subsequently for another 24 h for gross behavioural changes. Mortality was recorded for each group at the end of this period and the LD₅₀ of the mice were estimated as described by Lorke (1983).

Experimental animals

Twenty healthy male albino Wister rats of average weight 115 ± 2.8 g were used for the study. Animals were housed in stainless steel cages and acclimatized under standard environmental condition (27°C and 12 h light/dark cycle) with access to water and animal feed *ad libitum* for 10 days.

The animals were randomly divided into two groups of ten rats each and fed animal feed. The group 1 animals were given water only, while the group 2 animals were, with the aid of an intubator, orally administered 200 mg/kg aqueous extract of *L. owariensis* twice daily (morning and evening) for 10 days. At the end of the experimental period, the animals in each group were anaesthetized with chloroform and blood was collected by cardiac puncture. The blood sample from each animal was divided into two and dispensed respectively into a plain container and an EDTA bottle. Blood in the plain container was allowed to clot and serum separated by centrifugation at $600 \times g$ for 15 min. The serum was used for the liver function test. The EDTA anti-coagulated blood was used for the determination of haemoglobin concentration.

Biochemical analysis

The activities of alanine amino transferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP) were determined by standard methods as described by Balistreri and Shaw (1987).Total protein, albumin and bilirubin assay were determined using a chemistry analyzer (Ciba-Corning 550 Express Plus, USA). The haemoglobin concentration was estimated using the cyanmethaemoglobin method as described by Dacie and Lewis (1994).

Statistical analysis

Data obtained were expressed as mean \pm standard deviation and analyzed using student's 't' test. Values for P \leq 0.05 were taken to be statistically significant (Parker, 1979).

RESULTS AND DISCUSSION

Preliminary phytochemical screening of the leaves revealed the presence of alkaloids, flavonoids, tannins and saponins in the aqueous extract of the plants (Table 1). These secondary plant metabolites perform several functions in plants and may exhibit different biochemical and pharmaceutical actions in animals when ingested, and even in micro-organisms upon exposure. Such actions range from cell toxicity to cell protective effects (Trease and Evans, 1996).

The acute toxicity studies produced an LD₅₀ of 3370 mg/kg body weights of experimental mice. Table 2 indicates that the aqueous extract of L. owariensis does not have deleterious effects. Although, there were slightly higher but non-significant increase in the activities of ALT, AST and ALP in the serum of the treated animals compared to the controls. The result can be attributed to increase in hepatocellular activity given the observed significant reduction in total bilirubin concentration, which may be due to increased albumin synthesis and bilirubin conjugation (Balistreri and Shaw, 1987). Thus, the reduction in total bilirubin concentration may be attributed to the increased albumin levels since albumin transports bilirubin to liver for conjugation and subsequent excretion. This suggested hepatocellular function-enhancing effect of the extract may result from the action of the various

Group	Total Protein (g/l)	Albumin (g/l)	Total Biliribin (μmol/l)	Alkaline phosphatase IU/L)	Alanine amino Transferase (IU/L)	Aspartate amino transferase (IU/L)	Haemoglobin (g/l)
Control	53.0±2.4	31.0±1.1	160.7±1.2	95.0±2.5	78.0±6.7	144.0±7.2	137.0±1.6
Extract Treated Rats	51.0±2.6	32.0±1.5	145.4±1.6	97.0±2.6	80.0±5.6	146.0±5.8	115.0±1.2
t-value	1.69	1.59	22.84	1.65	0.68	0.65	10.78
Р	>0.05	>0.05	< 0.05	>0.05	>0.05	>0.05	<0.05

Table 2. Effects of aqueous extract of Landolphia owariensis on biochemical parameters of albino rats.

contents of the extract especially the presence of flavornoids which have been reported to have antioxidative effects (Middleton Jr., 1996). Furthermore, saponins also present in the extract are known to have hypocholesterolemic activities (Price et al., 1987), which may aid in lessening the metabolic burden on the liver.

In addition, saponins which have been found to be the most abundant constituent of leaves of medicinal plants and vegetables, exhibit appreciable anti-microbial and anti-inflammatory activities (Ebi and Ofoefule, 1997; Owoyele et al., 2002). This may translate to less need for and thus reduced synthesis of globulins especially immunoglobulins and may explain why, with increase in albumin concentration, the total protein concentration instead of increasing, decreased in the extract-treated rats (Table 2). In the same vein, tannins also detected in the extract are complex phenolic polymers which can bind to proteins and carbohydrates resulting in reduction in digestibility of these macromolecules and on the other hand, inhibition of microbial growth. Thus, the presence of tannins in the extract indicates that the plant extract has anti-microbial potential (Bulter, 1989), as has been shown in our previous studies on L. owariensis as an anti-microbial agent (Nwaogu et al., 2007).

The extract was also noted to reduce the haemoglobin concentration of test animals (Table 2) . The observation corroborates the report of Oluwole and Balarinwo (1997) that methanolic extract of some plants such as *Tatropha curcus* cause drastic reduction in haemoglobin concentration and other haematological parameters in experimental rats. This effect may be attributed to the presence of alkaloids in the leaf extract of *L. owariensis*. Alkaloids are basic natural products occurring primarily in many plants. They are generally found in the form of salts with organic acids, are haemolytically active and are also toxic to micro-organisms (Cheeke, 1989).

The results of the present study indicate that aqueous extract of *L. owariensis* administered orally at 200 mg/kg does not have hepatotoxic effects in rats. The reduction in haemoglobin concentration may not be attributed to intravascular haemolysis since the total bilirubin concentration of the treated animals decreased with administration of the extract. However, this calls for more research to ascertain this haemoglobin lowering-effect and to also explain the process of activity of the extract. It may also be interesting to study the possible effects of lower concentration of the plant extract on haemoglobin levels.

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