Effects of Aqueous Ethanolic Leaf Extract of *Boscia* senegalensis (PERS) on Liver Function of Wister Strain Albino Rats

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ABSTRACT

Boscia senegalensis has long been used as food, pesticide and medicine for the elimination of intestinal parasite from camel. The objective of this study was to evaluate the acute and sub acute toxicity (21 days) of *Boscia senegalensis* on the liver of Wister strain albino rats of both sexes via the oral route. For the acute toxicity, *Boscia senegalensis* extract was administered in a dose of 1000mg/Kg and 2000mg/Kg/day (n=3) while in the sub acute toxicity test, the following doses were administered; 100mg/Kg, 150mgKg, 200mg/Kg/day (n=5/group) for 21 consecutive days. In the acute toxicity test, *Boscia senegalensis* produce some toxic signs with no mortality. During the sub acute treatment with *Boscia senegalensis*, ALT was significantly high at P<0.01. However, Albumin and total protein were not significant at P>0.05. Therefore, the acute and sub acute administration of the aqueous ethanolic extract of *Boscia senegalensis* produced a toxic effect on Wister strain rats which is proven statistically to be dose dependent.

Keywords: Boscia senegalensis, wister strain albino rats, liver function test

Introduction

Toxicity is the ability of a chemical to damage an organ system, such as the liver or kidney, or to disrupt a biochemical process, such as the blood forming mechanism, or to disturb an enzyme system at some site in the body. It can be defined simple as the property of a chemical which causes damage to the body of a living organism (Yakubu *et al*, 2003).

It is classified into acute, sub acute or chronic based on the rate of onset symptoms, and the duration of exposure to the offending agent. Acute toxicity is usually produced by the single or sudden intake of a substance in quantity large enough to cause severe depression of a vital physiologic function.

Sub acute toxicity differs from acute toxicity only with respect to the conditions under which the subject is endangered by the drug or extract. In sub-acute toxicity, there is frequent, repeated exposure over several hours or days to a dose insufficient to produce deleterious effects when given as a single dose (Seck, 1993).

The advancement in toxicology and also molecular biology and biochemistry is paving the way for the paramount development in the estimation of the dangers posed by the numerous amounts of substances (chemicals) found in the low levels in our surrounding (Orwa *et al*, 2009)

Toxicological analysis is approaching a point where it uses the advantage of the revolution in biology and biotechnology. The new process is the product of systems that have addressed advancement in science through the increments in test protocols and through the addition of new test materials.

More so, the acquired data may not answer the question concerning risk to human health. However, the U.S Environment Protection Agency (EPA) concluded that it is time for a more innovative approach to toxicity testing and they employed the National Research Council (NRC) to develop durable vision and strategy for toxicity testing (Committee on Toxicity testing and Assessment of environment Agents, 2007).

Materials and Methods

Test materials

Aqueous ethanolic (70:30 ethanol: water) leaf extract of *Boscia senegalensis* (pers) was used in the investigation. *Boscia senegalensis* used for this research work was collected in November on the bank of the river Yobe in Geidam, Northeastern Nigeria and was identified by the Taxonomy unit of the Biological Sciences Department, University of Maiduguri, Nigeria. The dried extract was stored in a desiccator. The material was stored in a refrigerator at 4^oC and protected from light until time of drug administration when appropriate stock solutions were made in distilled water for administration to the experimental animals.

Che micals

Kits for Aspartate amino transferase, Alanine amino transferase, alkaline phosphatase, Total protein and Albumin were supplied by Randox.

Acute Toxicity studies

The acute toxicity (LD_{50}) was estimated orally in rats following Lorke's method (1983). Dose levels used ranged from 1000mg/kg to 2000mg/kg. Clinical signs in each group of rats (within 72h) were monitored.

Sub Acute Toxicity

Twenty Wister rats were divided into four groups of five rats each. No extract was administered to the first group. Other three groups were given 100mg/kg, 150mg/kg and 200mg/kg for the period of 21 days, during which observations were made and their weight noted weekly. The animals were sacrificed on the 22nd day without food or extract administration.

However, during the period of the 21 days, the animals were fed *ad libitum* with normal feed and water. The blood sample was collected. (No EDTA was added).

Preparation of sera samples

On Day 21 of the dosing, all the animals were sacrificed and the blood samples were collected from the animal. The sample were collected in plain plastic tubes and allowed to stand for 3h to ensure complete clotting. The clotted blood samples were centrifuged at 3000rpm for 10mins and clear serum sample were aspirated off and stored frozen.

Serum biochemistry

The following parameters were determined colorimetrically by employing the standard readyto-use kits of Randox(RANDOX Laboratories Ltd., 55 Diamond Road, Crumlin, Co. Antrim, United Kingdom. BT29 4QY): Aspartate amino transferase, Alanine amino transferase, Alkaline phosphatase, Total protein and Albumin. The manufacturer's instructions for each biochemical parameter were strictly followed in the course of the investigations.

Statistical Analysis

All data were expressed as Mean±SEM. Results were statistically analyzed by student's t test for significant differences between group means. 95% level of significance (p<0.05), 99% level of significance (p<0.01) and 99.9% level of significance (p<0.001) were used for the statistical analysis.

RESULTS

Acute toxicity studies

At the dose levels tested, these clinical signs were observed; the animals were weak, their furs were erected, their eyes were red, and there was a general loss of appetite. No mortality was observed in the different groups of rats that receive *Boscia senegalensis* orally after 72 hours. Hence the LD₅₀value was estimated to be >2000mg/kg body wt.

Clinical signs and mortality patterns

During the 21 days of treatment, a mild sedative effects, anorexia and respiratory distress were observed, however no other advert clinical manifestation (e.g. diarrhoea, haematuria, restlessness, uncoordinated muscles movements etc) were perceptible in the experimental animals during the dosing period

 Table 1: Some changes in liver biochemical indices following Boscia senegalensis administration for 21 days.

Group A	Group B	Group C	Group D
(Control)	(100mg/kg)	(150mg/kg)	(200mg/kg)
ALT(Iu/l) 19.4±0.98	27.4 ±0.98***	29.2 ±0.86***	30.0 ±1.00***
AST(Iu/l) 62.2 ±6.88	77.8 ±1.11*	79.2 ±2.48*	87.2 ±4.63**
ALP(Iu/l) 84.2 ±4.96	92.2 ±2.00 ^{ns}	$101.2 \pm 0.2 **$	102.4± 0.24**
ALB(g/l) 31.2 ± 1.32	30.4 ± 1.14^{ns}	31.7 ± 0.73 ^{ns}	30.2 ± 0.73 ^{ns}
TP(g/l) 72.4 ± 2.02	71.0 ± 0.55 ^{ns}	72.6 ± 1.78 ^{ns}	$70.6 \pm 0.60^{\text{ ns}}$

Values are express as Mean±SEM

^{ns}P>0.05, not significantly different from control rats(Student t-test)

* P<0.05, significantly different from control rats (Student t-test)

** P<0.0, significantly different from control rats (Student t-test)

*** P<0.001, significantly different from control rats (Student t-test)

ALT- Alanine aminotransferase, AST- Aspartate aminotransferase, ALP- Alkaline phosphatase, ALB-Albumin, TP- Total protein.

DISCUSSION

Some plant products show systemic toxicity after the chemical experimental treatment, what can be characterized by the reduction in the animals' body mass, behavior, hematological and biochemical alterations (John and Gunzel, 1997). The toxic risks are due to the substances found in the plant which can reduce liver and kidney toxicity. (Cavalli *et al*, 2004) The alkaline phosphatase, albumin, ALT, AST and total protein level are given in table 1. Using the student 't' test to compare between B(100 mg/dl) and normal, C (150 \text{ mg/dl}) and normal, D (200 \text{ mg/dl}) and normal. The value of alkaline phasphatase in group C(150 \text{ mg/dl}) and the value in group D(200 \text{ mg/dl}) when compared with the normal (A), shows that there is significant increase at P<0.01. The toxic effect caused by the extract may be due to the destruction of the hepatic cell membrane by the extract (John and Gunzel 1997). However, there was no significant increase or decrease when group B(100mg/dl) is compared to group A, P > 0.05. The albumin and total protein level at all the doses administered shows no significant change at P>0.05.

Nevertheless, ALT significantly increased at P<0.001 for all the doses administered. AST significantly increased at P<0.05 for group B and C. It significantly increase at P<0.01 for group D. The toxic effect caused by the extract may be due to the destruction of the hepatic cell membrane by the extract. Further statistical analysis to check if the toxicity was dose dependent or not indicate that toxicity is highly dependent on dosage.

Toxic effect observed is characterized by the destruction of the cell membrane of hepatic cells by the component of the extract. (Cavalli *et al.*, 2004) The destruction of the hepatic cells by the components of *Boscia senegalensis* lead to the release of the marker enzymes (ALT, AST, ALP) into the blood.

Therefore, this research work shows that the ethanolic leaf extract of *Boscia senegalensis* consumed by most destitute areas in West Africa increases the ALT, AST and ALP level in the liver. Therefore the plant leaf is hepatotoxic.

The leaves of *Boscia senegalensis* should not be used as food because of the toxic activity on the liver as shown in the result. However, it should be used in Agriculture as parasiticide, in food storage (granary) and removal of intestinal parasite in Camel and other hoof animals.

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