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Full Length Research paper

Serum biochemical response of rats fed with *Sclerocarya birrea* juice extracts

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Sclerocarya birrea **fruits are widely eaten in developing countries especially in rural areas and serve as nutrients supplements. However, they also contain phyto-toxin which may affect the normal functioning of the body. The present study examined the effect of feeding albino rats with *S. birrea* juice with respect to their body weight, liver and kidney biochemical parameters. The results showed that no mortality was recorded in acute toxicity (LD_{50}) study. Rats fed with 1000, 2000 and 3000 mg/kg body weight experienced an increase in the body weight through out the period of treatment compared with the initial body weight, while those fed with 4000 mg/kg experienced a decrease in the body weight compared with the initial body weight. In biochemical response, there was a significant ($p<0.05$) increase in serum albumin, total bilirubin in rats fed with 3000 and 4000 mg/kg body weight. Similarly, serum enzymes activities, aspartate amino transferase (AST), serum alanine amino tranasferase (ALT) and alkaline phosphatase (ALP) were significantly ($p<0.05$) elevated at higher doses (3000 and 4000 mg/kg) compared to the control group, which is an indication of organ toxicity by cellular destruction induced by phyto-toxin present in the juice. Renal function indices – serum creatinine, urea, uric acid and electrolytes in rats fed with 3000 and 4000 mg/kg body weight showed a significant ($p<0.05$) change compared to the control group. The results of this study showed that *S. birrea* juice has a relatively low or no toxicity profile.**

Key words: *Sclerocarya birrea*, albino rats, toxicity, extracts serum.

INTRODUCTION

A significant proportion of indigenous fruits in West African sub region are seasonal forest products harvested for consumption on site or transported to other areas particularly urban centers for sale (Nnam and Njoku, 2005). The knowledge of the nutrient composition of some of these fruits enhance their use and increase their consumption which in turn improves the nutrient profile of a good proportion of the populace (Nzeagwu and Onimawo, 2010). One of such trees is *Sclerocarya birrea* (Anacardiaceae) which its botany was well reported by Moganedi et al. (2007), Hillman et al. (2008) and Ojewole et al. (2010). The tree bears pale yellow fruits (Plate 1) with a plain tough peel and fibrous juicy sweet-sour mucilaginous flesh (Hillman et al., 2008). *S. birrea* fruits are widely eaten in developing countries not only during the period of food scarcity but during period of abundance; perhaps due to cultural acceptance (Ojewole et al., 2010).

Nutritional study of the plant's fruits revealed that the fruit juice contained 3.31% dry weight (DW) and 90.35% (DW) available carbohydrate (Hassan et al., 2010). Glew et al. (2004) and Moganedi et al. (2007) reported that *S. birrea* seed kernels are edible and rich in oil (50 to 60%) and protein (28 to 36%). On dry weight, *S. birrea* seed kernels contained appreciable amount of copper (24.8 µg/g),

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Plate 1. Ripe fruits of *S. birrea.*

magnesium (4210 µg/g) and zinc (62.4 µg/g) (Glew et al., 2004).

S. birrea tree was also reported to possess medicinal properties. Ojewole (2004) reported that *S. birrea* stem-bark aqueous extract is safe, and/or non-toxic to mice and possesses analgesic, anti-inflammatory and anti-diabetic properties. Ojewole et al. (2010) noted that polar extracts of *S. birrea* leaf and stem-bark (inner bark) have antibacterial and antifungal activities.

Even though wild plants are important sources of nutrients and phytocompounds that play a role in protecting against conditions such as cardiovascular disease and cancer, they also contain other compounds that may lead to hepatic/tubular necrosis (Caswell, 2009). This paper aimed at investigating the toxic effect of *S. birrea* fruit juice using albino rats as experimental animals.

MATERIALS AND METHODS

Sampling and sample treatment

2 kg of mature *S. birrea* fruits were collected in June, 2010 at More town, Kware local government of Sokoto State, Nigeria. The site was chosen because of the abundance trees of this plant. Five samples of the plant were randomly selected and the fruits were collected from different branches of the selected plant, as described by Hassan and Umar (2004). The sample was transported to the laboratory in polyethylene bags.

The samples were mixed together, thoroughly washed with distilled water and the residual moisture evaporated at room temperature. The juice was manually made by squeezing the fruits and used for the toxicity studies.

Preparation of juice extracts

100 cm^3 of the juice was placed in already weighed crucible and dried in an oven (Gallenkamp, England) at 50°C to a constant weight. The extract was cooled in a desiccator and weighed, the amount of extract was calculated in grammes per 100 cm^3, that is, (g/100 cm^3) (AOAC, 1990). The procedure was repeated to obtain more extract. The extract was then reconstituted in sterilized distilled water.

Toxicological studies

Animals

Albino rats (males and females) weighing 169 to 320 g were purchased from the Department of Biological Sciences, Usmanu Danfodiyo University, Sokoto, Nigeria. The animals were kept at the animal house of the department in a wire mesh cages. They were fed with grower's feed and tap water *Ad libitum* for two weeks to acclimatize before starting the experiments.

Administration of extracts

Acute toxicity studies (determination of LD_{50})

Aqueous extract of the juice (3000 mg/kg body weight) was administered to 5 groups of one rat each (one after the other at a grace observation period of 24 h) in a single oral dose using a feeding needle. The control group received equivalent amount of distilled water.

Observation for toxic symptoms was made and recorded systematically at 1, 2, 4 and 6 h after administration. Finally, the number of survivors was noted after 48 h for each animal.

The toxicological effect was assessed on the basis of mortality, which was expressed as LD_{50} and calculated using the limit test dose, up and down procedure of Organization for Economic and

Table 1. Weight of rats (g) as affected by doses of *S. birrea* juice (extracts) after four weeks.

Dose (mg/kg)	Initial weight	1st week	2nd week	3rd week	4th week
0.00(control)	169.56±6.88	170.27±9.56	178.10±4.33	189.77±2.35	174.37±5.74
1000	191.06±2.70	192.77±5.98	199.27±6.30	199.80±6.90	200.70±7.13
2000	216.57±6.51	217.80±9.90	219.47±7.32	219.81±5.00	220.16±5.62
3000	220.30±8.51	227.27±8.50	227.87±3.44	231.67±7.22	240.83±6.77
4000	320.10±2.11	319.00±4.11	318.40±1.22	314.50±0.12	313±3.80

Values are mean ± standard deviation.

Table 2. Liver function indices in rats administered with *S. birrea* juice (extracts).

Dose (mg/kg)	TOP (g/dl)	ALB (g/dl)	TB (g/dl)	ALP (U/L)	ALT (U/L))	AST (U/L)
0.00(control)	6.0±0.66	4.62±0.77	4.94±0.59	101.7±2.7	9.33±3.12	26.33±2.93
1000	5.99±0.50	3.87±0.21	5.06±0.77	319.2±2.9	13.67±0.58	35.0±3.46
2000	5.33±0.87	3.62±0.21	5.08±0.49	406.6±5.9	23.77±0.68	39.50±3.04
3000	5.32±0.38	3.50±0.43*	5.18±0.21*	427.8±2.0*	28.33±0.57*	54.23±3.28*
4000	5.24±0.24	3.13±0.37*	6.91±0.46*	453.6±3.0*	31.0±1.00*	59.03±0.21*

Values are mean ± standard deviation. TOP, total protein; ALB, albumin; TB = total bilirubin; ALP, alkaline phosphatase; ALT, alanine aminotransferase; AST, asphatate aminotransferase. * = Significantly different from the control ($P < 0.05$) using one way analysis of variance (6).

Cultural Development (OECD, 2001).

Sub-acute toxicity studies

A total of thirty albino rats were divided into five groups of six rats each. Animals in Groups 2, 3, 4 and 5 were orally administered with sample extract 1000, 2000, 3000 and 4000 mg/kg body weight once daily for 28 days respectively. Animals in Group 1 served as the control group (that is, 0.00 mg/kg) and received only drinking water by the same route. The body weights of all the animals before and within 28 days (weekly) of treatment were recorded.

Blood sample and clinical chemistry

The animals were sacrificed 24 h after the last treatment; blood samples were collected, allowed to clot and then centrifuged at 3000 rpm for 10 min to obtain sera. The biochemical parameters, serum total protein (TP) and total albumin (TA) were determined by the method of Cheesbrough (1991). Total bilirubin (TB) was analyzed (Randox kit) using the method reported by Hassan et al. (2005). Serum alanine amino tranasferase (ALT) and aspartate amino transferase (AST) were determined using Randox assay kit by standard methods of Reiman and Frankel (1957). Alkaline phosphatase (ALP) was estimated by the Randox (colorimetric) method of Rec (1972). Serum electrolytes and creatinine (colorimetric with deproteinization) were estimated by the methods of Henry (1974). Urea (diacetylmonoxime) was analyzed using the method of Wybenga et al. (1971). Uric acid was estimated using the method of Morin and Prox (1973).

RESULTS AND DISCUSSION

Body weight

There was a noticeable increase in the body weight of the animals that received 1000, 2000 and 3000 mg/kg of the extracts compared with the initial body weight of the animal. There is also a noticeable decrease in the body weight of the animal that received 4000 m/kg of the extracts compared with the initial body weight of the animal (Table 1).

Acute toxicity (LD_{50})

Acute toxicity at 3000 mg/kg body weight of the juice extract produced no mortality after 48 h of observation, indicating that the mean (LD_{50}) of the extract is greater than 3000 mg/kg body weight.

Subacute toxicity

There was a significant ($p<0.05$) change in the liver function indices (Table 2) and kidney function indices (Table 3) at higher doses of the extract compared with the control.

The percentage yield

The percentage yield of the extract was 12.32 g/100 g of the juice which is an indication that the juice could contain some important nutritional or medicinal phytocompounds.

Acute toxicity (LD_{50})

Acute toxicity test at 3000 mg/100 kg of *S. birrea* produced no mortality after 48 h of observation which

Table 3. Kidney function indices in rats administered with *S. birrea* juice (extracts).

Dose (mg/kg)	Creatinine (µmol/L)	Urea (Mmol/L)	Uric acid (µmol/L)	Sodium (ppm)	Potassium (ppm)
0.00(Control)	81.42±0.10	9.96±0.00	201.20±2.20	31.30±3.12	8.32±2.10
1000	88.50±0.20	10.71±1.16	205.10±4.10	31.06±0.11	8.71±0.40
2000	90.06±0.01	11.34±1.54	212.20±5.00	30.00±0.10	8.82±1.20
3000	97.35±0.03*	13.94±0.57*	216.40±1.20*	28.07±0.11*	8.90±0.80*
4000	104.96±0.10*	14.11±0.55*	220.01±2.20*	28.00±0.10*	9.80±2.60*

Values are mean ± standard deviation. *= Significantly different from the control ($P < 0.05$) using one way analysis of variance (6).

indicates that the mean lethal dose (LD_{50}) of the juice extract is greater than 3000 mg/100 kg body weight. Generally, acute toxicity did not produce any grossly negative behavioural changes such as excitement, restlessness, convulsions or coma in the rats.

Sub acute toxicity

In sub chronic toxicity (Table 1), it was observed that the *S. birrea* fruit juice (extract) caused a decrease in the body weight of rats administered dose of 4000 mg/kg at the 1st, 2nd, 3rd and 4th week of treatment. The reduction in weight could be due to reduced fluid and water intake, which may be secondary to a feeling of fullness and loss of appetite after administration of the juice extract (Joseph et al., 1989). The reduction in the body weight could be due to high tannins content which might hinder protein bioavailability (Umar, 2008).

The result of liver function indices was presented in Table 2 and there was no significant change ($P < 0.05$) in the serum total protein of the rats administered with different doses of the juice extracts, but there was a significant ($P < 0.05$) decrease in the serum total albumin of the rats administered dose 2000, 3000 and 4000 mg/kg body weight. Albumin is synthesized by the liver and as such, it represents a major synthetic protein and is a marker of the ability of the liver to synthesize proteins (Johnston, 1999). The decrease in the serum albumin indicates that the synthetic function of the liver has been affected though malnutrition can also cause decrease in albumin (hypo albuminemia) without associated liver disease. A significant ($P < 0.05$) decrease in the serum albumin clearly shows that the juice extract inhibits protein synthesis in the rats although the values are still within the normal range of (5.6 to 7.6 g/dl) as reported by The Rat Fan Club (2010).

Bilirubin is a major break down product of haemoglobin (Oboh, 2005). The water solubility of bilirubin allows the bilirubin to be excreted in the bile; the bile is then used to digest food.

As the liver becomes irritated, the total bilirubin may rise. As presented in Table 2, there was a significant ($P < 0.05$) increase in the total bilirubin in the serum of rats administered dose of 3000 and 4000 mg/kg body weight which is an indication that the juice extract interfere with the metabolism of bilirubin in the liver (Oboh, 2005).

As presented in Table 2, there was a significant ($P < 0.05$) increase in the serum ALP of rats administered with the juice extract. ALP is a marker enzyme for the plasma membrane and endoplasmic reticulum (Wright and Plummer, 1974). The significant ($P < 0.05$) increase in the serum ALP could be due to renal or intestinal damage, biliary tract damage and inflammation (Oboh, 2005). More so, the increase could be attributed to enzyme activation by the phytochemical constituents of the *S. birrea* juice.

The ALT and AST are liver specific enzyme markers of necrotic injury and cholestasis (Speech and Liehr, 1983). The result obtained (Table 2) shows that there is a significant ($P < 0.05$) increase in the serum ALT and AST of the rats administered dose of 3000 and 4000 mg/kg body weight. The significant increase could be due to damage to the hepatitic cell or heart attack (Hearly et al., 1995) and may have been induced by some phytocompouds of the juice extract.

Serum urea, uric acid, creatinine and electrolytes are markers of damage to renal function (Harold et al., 1980). As presented in Table 3, aqueous juice extract of *S. birrea* fruit at 3000 and 4000 mg/kg body weight induced a significant ($P < 0.05$) increase in kidney function indices and may interfere with renal functions. The significant ($P < 0.05$) decrease in sodium and increase in potassium in the group treated with 3000 and 4000 mg/kg body weight are also signs of renal failure (Hassan et al., 2005). The changes in biochemical indices of renal function may have been induced by the phytochemical constituents of the juice extract.

Conclusion

The result indicated a significant ($p < 0.05$) decrease in body weight of rats at high dose (4000 mg/kg body weight) compared to control group which could be due to the presence of antinutritional factors such as tannins in the juice. The biochemical parameters indicated that the juice has effect on liver and kidney functions at high dose. From the results, *S. birrea* fruit juice has a relatively low toxicity at high dose and for that the juice should be

used cautiously and in small doses.

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