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The Effects of *Boswellia dalzielii* (Burseraceae) Aqueous Bark Extract on Rat Liver Function

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Abstract: The effect of the aqueous bark extract of *Boswellia dalzielii* (Burseraceae) on liver function was investigated using female Wistar rats. Graded concentrations of 0, 50, 100, 150 and 200 mg kg⁻¹ body weight, of the aqueous bark extract of the plant sample were administered to the rats through gastric route and alkaline phosphatase, aspartate aminotransferase and alanine aminotransferase activities, total protein, albumin and bilirubin (total, conjugated and unconjugated) were determined in the serum. Phytochemical screening of the extract was also done. The extract of *B. dalzielii* was found to have no significant ($p > 0.05$) effect on total protein and albumin content at the end of the 5 days treatment period. Total bilirubin and alkaline phosphatase activities did not show any significant ($p > 0.05$) difference as compared with the control. However, unconjugated bilirubin was found to decrease significantly ($p < 0.05$) with increased dose of extract. These results seem to suggest no damage to hepatocytes as a result of the exposure to the plant bark extract. Moreover the transaminase activities seem to corroborate this, since they decreased significantly ($p < 0.001$) with increase in the concentration of extract, as compared to control values, suggesting that *B. dalzielii* bark extract may strengthen the liver. The presence of flavonoids further gives credence to the hepato-strengthening activity of *B. dalzielii* bark extract.

Key words: *Boswellia dalzielii*, hepato-strengthening, transaminases, bilirubin, flavonoids

INTRODUCTION

Boswellia dalzielii (Burseraceae) is a tree plant, abundantly found in north-western Nigeria, where the Hausa speaking people refer to it as *Hano* or *Harrabi*. This plant is very popular among the locals as a potent source of ethnomedicine. The extract from its leaves is used for the treatment of diarrhea in poultry. The root decoction of *B. dalzielii* and *Daniella oliveri* is used for wound healing (Etuk *et al.*, 2006a). The fresh bark is eaten to induce vomiting and relieve symptoms of giddiness and palpitations. The root decoction of the plant boiled along with *Hibiscus sabdariffa* is used for the treatment of syphilis. The fragrant gum resin from the plant is used locally for fumigation of clothes and houses and as a deodorant (Etuk *et al.*, 2006a). Oil from the leaves of *B. dalzielii* was found to exhibit significant activity against *S. aureus*, *B. subtilis* and *C. albicans* (Nwinyi *et al.*, 2004). Also, the aqueous stem bark extract of *B. dalzielii* was reported to show anti-ulcer activity and reduced gastrointestinal motility (Nwinyi *et al.*, 2004) and to possess anti-diarrhoeal effect, which may be related to anticholinergic mechanisms (Etuk *et al.*, 2006b). Crude extracts of the stem bark of this plant

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have been found to show antibacterial activity against both Gram-positive and Gram-negative bacteria (Olukemi *et al.*, 2005). Despite the widespread uses of this plant in treating a plethora of human and animal diseases in this environment, little work has been done on its phytochemistry and its effects on some major organs in the body.

This study was undertaken to investigate the effects of the aqueous bark extract of *B. dalzielii* on the liver function. This was with a view to assessing the possible cytotoxic side effect that might arise as a result of consumption of this plant extract, since liver is the major organ of drug metabolism.

MATERIALS AND METHODS

Collection of Plant Sample

The stem bark of *Boswellia dalzielii* was collected from Unguwan Hakimi-Dengi, Plateau State, Nigeria, during the dry season (in the month of January 2004). The plant was identified by Ibrahim Muazzam, a plant taxonomist in the Department of Medicinal Plant Research and Traditional Medicine, National Institute for Pharmaceutical Research and Development (NIPRD), Abuja, Nigeria. A voucher specimen (No. 4592) is deposited at NIPRD herbarium.

Preparation of Plant Extract

One gram of the dried pulverised bark of *B. dalzielii* was steeped in 100 mL of hot distilled water (60°C) and kept at room temperature for 30 min with occasional stirring. Thereafter, the mixture was filtered using Whatman No. 1 filter paper and the filtrate (extract) was transferred into dark-brown bottle and used soon after. The extract was prepared fresh each time required. The actual amount of solubles extracted from 1 g of the sample was determined after evaporating the solvent and weighing the residue (462.5 mg g⁻¹).

Experimental Animals

Thirty female Wistar albino rats, between 7 and 8 weeks old, weighing between 180 and 200 g, were obtained from the Small Laboratory Animal House, National Institute for Trypanosomiasis Research (NITR), Vom, Jos and transferred to the University of Jos Animal House where they were allowed to acclimatise for 7 days and where the work was carried out. They were maintained in the experimental facility, provided standard feed for laboratory rodents and regular tap water *ad libitum*. Six animals were housed together until the commencement of the experiment.

Treatment of Animals

Five groups (A-E) of 6 rats each were used for the study. Group A (control group) did not receive any dose of the extract. Group B received the normal, recommended daily dose of 50 mg kg⁻¹ body weight (extrapolated from human dose), whereas groups C, D and E received 100, 150 and 200 mg kg⁻¹, respectively, through gastric intubation, for 5 consecutive days. Six hours after the last dose of the extract, all the animals in the five groups were sacrificed by decapitation. Blood from each animal was collected separately in labelled centrifuge tubes. They were spun at 5,000 x g for 15 min using MSE Minor centrifuge at room temperature. The serum samples were obtained and stored at -4°C in labelled sample bottles until required.

Quantitative Estimation of Serum Total Protein

Serum total protein was determined by the biuret method, as described by Gornall *et al.* (1949).

Serum Albumin Determination

Serum albumin concentration was determined by the bromocresol green (BCG) binding method, as described by Cheesbrough (1991).

Serum Bilirubin Determination

The serum total bilirubin was estimated by colorimetric method based on that developed by Jendrassik and Grof, as described by Willard and Meites (1982). Conjugated bilirubin was determined by the direct Van den Bergh's reaction. Unconjugated bilirubin was determined by subtracting conjugated from total bilirubin.

Measurement of Serum Alkaline Phosphatase (ALP) Activity

The serum activity of ALP was determined by the Bessey Lowry and Brock method, as described by Cheesbrough (1991).

Determination of Serum Aspartate Aminotransferase (AST) Activity

The serum activity of AST was determined by the Reitman-Frankel method, as described by Cheesbrough (1991).

Determination of Serum Alanine Aminotransferase (ALT) Activity

The serum activity of ALT was determined by the Reitman-Frankel method, as described by Cheesbrough (1991).

Phytochemical Analysis

The phytochemical screening of the bark extract was carried out using standard qualitative procedures as described by Trease and Evans (1989) and Sofowora (1993). The extract was screened for alkaloids (Mayer's test and Dragendorff's test), flavonoids (NaOH test, FeCl₃ test and lead acetate test), glycosides (Salkowski test and Keller-Killiani test), saponins (Frothing test), tannins (FeCl₃ test), steroids and triterpenoids (Lieberman-Burchard test).

Statistical Analysis

Statistical analysis of data was performed using Stat-view for Mackintosh software programme (Release 7.0). Group comparisons were done using the analysis of variance (ANOVA). A p-value of <0.05 was considered statistically significant.

Ethics

This study was carried out with respect for the welfare of animals, as recommended by WHO (1992). Moreover, all procedures involving animals were carried out in strict compliance with the Animal Ethics Committee rules and regulations of the University.

RESULTS

The effect of *Boswellia dalzielii* bark extract on the serum levels of total protein, albumin, bilirubin, alkaline phosphatase and transaminases (AST, ALT) are given in Table 1. A slight decrease was observed in serum total protein concentration, but this was not statistically significant ($p > 0.05$). The administration of extract did not have any significant ($p > 0.05$) effect on the albumin component of serum.

Boswellia dalzielii extract had no detectable effect on the conjugated bilirubin. However, it did have significant ($p < 0.05$) effects on the total and the unconjugated bilirubin, at high doses (150 and 200 mg kg⁻¹). The extract did not have any significant ($p > 0.05$) effect on ALP activity in general. However, at 150 mg kg⁻¹ ALP activity showed a decrease, which still was not statistically significant ($p > 0.05$). There were significant ($p < 0.001$) decreases in the activities of serum transaminases (AST and ALT) in the treated groups as compared to the control group. This effect appeared to be dose dependent.

Table 1: Effect of *Boswellia dalzielii* bark extract on some biochemical parameters of liver function test

Treatments	Total protein (g/100 mL)	Albumin (g/100 mL)	Bilirubin (mg/100 mL)			Alkaline phosphatase (LU)	Transaminases (U L ⁻¹)	
			Conjugated	Unconjugated	Total		AST	ALT
Control	9.70±1.49	4.24±0.10	0.09±0.16	0.22±0.08	0.31±0.02	58.47±25.06	7.63±0.26	10.21±0.03
50 mg kg ⁻¹	7.83±2.16	4.24±0.10	0.18±0.08	0.14±0.01	0.31±0.08	61.26±28.48	6.49±0.09 ^b	10.08±1.38
100 mg kg ⁻¹	8.37±1.67	4.24±0.10	0.09±0.08	0.14±0.01	0.22±0.08	87.74±04.18	6.30±0.16 ^b	9.03±0.14 ^b
150 mg kg ⁻¹	7.97±2.60	4.18±0.00	0.04±0.08	0.09±0.08	0.13±0.14 ^a	38.98±08.69	6.22±0.10 ^b	8.90±0.12 ^b
200 mg kg ⁻¹	7.80±0.69	4.06±0.10	0.13±0.14	0.00±0.00 ^a	0.13±0.14 ^a	78.51±23.53	3.89±0.61 ^b	5.24±0.99 ^b

Tabulated values are mean±SD of six determinations, a p<0.05 vs control, b p<0.001 vs control

Table 2: Phytochemical composition of *Boswellia dalzielii* bark extract

Phytochemical Occurrence	Glycosides			
	Alkaloids	Cyanogenic	Digitalis	Cardiac
	+	-	-	-

Table 2: Continued

Flavonoids	Saponins	Tannins		Triterpenoids	Steroids
		Hydrolyzable	Pseudotannins		
+	+	-	+	-	-

+ Detected, - Not detected

The result of phytochemical analysis of *B. dalzielii* bark extract is as shown in Table 2. As can be seen from the table, glycosides, triterpenoids, steroids and hydrolysable tannins were not detected, while saponins, flavonoids, alkaloids and pseudotannins were detected in the plant bark extract.

DISCUSSION

Generally, a significant increase in serum or plasma total protein is an indication of tissue injury (Emerson *et al.*, 1993). In the present study, a slight decrease was observed in serum total protein concentration, following administration of *Boswellia dalzielii* extract, but this was not statistically significant. This decrease may, however, suggest that the extract had rather a protective effect on tissues.

A rise in the level of bilirubin in the blood can be as a result of overproduction of bilirubin caused by an excessive breakdown of red blood cells; the bilirubin being of the unconjugated type (Awad, 1997). Also, a rise in the blood total bilirubin level can be caused by liver cell damage (Cheesbrough, 1991; Awad, 1997). In this study, a marked decrease in total and unconjugated bilirubin levels was observed, suggesting that the extract may have a protective effect on both the red blood cells and the liver. Moreover, a progressive decrease was observed in unconjugated bilirubin concentration as the dose of extract increased. Also, at the dose of 200 mg kg⁻¹ unconjugated was completely converted into conjugated bilirubin, i.e., the concentration of conjugated became equal to that of the total bilirubin. These data suggest that the extract may enhance the conjugation of bilirubin in the liver.

The presence of flavonoids further gives credence to the hepato-strengthening effect of *Boswellia dalzielii* extract. In fact, natural flavonoids and polyphenolic compounds have been reported to exhibit protective and strengthening activities on liver cells (Adzet *et al.*, 1987; Akamatsu *et al.*, 2004; Oh *et al.*, 2004).

Whenever there is liver cell damage the serum or plasma levels of AST and ALT rise. In general, the higher the activities of both enzymes the greater the degree of liver damage (Cheesbrough, 1991; Gatsing *et al.*, 2005; Garba *et al.*, 2006). The decrease in the activities of these transaminases (AST and ALT) observed in the study corroborates the serum total protein and bilirubin contents, thus suggesting the strengthening action of the extract on the liver and other organs having significant quantities of these enzymes.

In the light of the foregoing, it appears that *Boswellia dalzielii* aqueous bark extract, at the doses used, may rather significantly strengthen the liver. This plant extract may therefore be used in the management of hepatic disorders. However, Etuk *et al.* (2006a) reported significant reductions in the packed cell volume and red blood cell count and significant increase in the serum urea level of the rats treated with a high dose (2700 mg kg^{-1}) of the aqueous stem bark extract of *Boswellia dalzeilii*. These findings suggest that prolonged oral administration of very high doses of the aqueous stem bark extract of *Boswellia dalzielii* may be associated with increased risk of toxicity.

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