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Effect of Aqueous Stem Bark Extract of Shea Butter (*Vitellaria Paradoxa)* Tree on Serum Glucose and Kidney Function Biomakers in Alloxan–Induced Diabetic Rats

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Plants are the sources of many important drugs of the modern world. This study is aimed at assessing the safety status of aqueous stem bark extract of Shea butter tree (*Vitellaria paradoxa)* as well as its effect on serum glucose and kidney biomarkers in alloxan-induced diabetic rats. Adult mice were administered oral doses, up to 5000 mg per kg body weight of the extract for toxicity assessment. Adult (male and female) albino rats weighing between 80-120 g were used for the study. The animals were divided into four groups of five (5) albino rats each as follows: Group A – Normal control (distilled water), Group B – Diabetic rats untreated, Group C – Diabetic rats treated with 100 mg/kg of aqueous extract of shea butter and Group D – Diabetic rats treated with 0.5 mg/kg anti-diabetic drug (Glibenclamide). Diabetes was induced by a single subcutaneous administration of alloxan hydrate (150 mg/kg body weight). Diabetic rats were treated with 100 mg/mL aqueous extract of the plant per kilogram body weight for two weeks. On the last day of treatment, the animals were anaesthetized, and blood samples were collected. The serums obtained were used for the measurement of glucose, urea, creatinine and sodium levels using standard methods. From the results, no mortality was observed at a dose less than or equal to 5000 mg/kg body weight. Treatment with the extract caused a significant ($p<0.05$) decrease in the blood glucose level and significantly normalizes blood urea, creatinine and sodium levels of alloxan-induced diabetic rats. Therefore, the extract may serve as a useful material for screening for antidiabetic drug.

Keywords: Shea butter tree, Serum glucose, Kidney function, Biomarkers

1. Introduction

Medicinal plants are the sources of many important drugs of the modern world. These plants contain substances that can be used for therapeutic purposes, and which are precursors for the synthesis of modern drugs (Kumar and Clark, 2002). Shea butter is an off-white or ivory-coloured fat extracted from the nut of African Shea tree (*Vitellaria paradoxa* formally *Butryspermum paradoxum*) (Alfred, 2002).

Shea tree is a plant that is locally abundant in Nigeria in the dried savannah zones, particularly near towns and villages (Ndukwe *et al*., 2007). Different parts of the plant including leaves, roots, seeds, fruit and stem bark have been used traditionally in the treatment of enteric infections such as diarrhea, dysentery, helminthes and other gastrointestinal tract infections, skin diseases and wound infections. In line with the need for more effective and safe anti-diabetic drugs, and to justify the traditional use of shea butter tree (*Vitellaria paradoxa)* as a remedy for the treatment of diabetes, this study is designed to assess the safety status of aqueous stem bark extract of Shea tree (*Vitellaria paradoxa)* as well as its effect on serum glucose and kidney biomarkers in alloxan-induced diabetic rats.

2. Materials and Methods

2.1 Collection of Sample

The plant sample (shea butter) was collected from Geben Damu of Kware local government area and authenticated at the Department of biological sciences Usman Danfodiyo University, Sokoto, Nigeria with identification number UDUH/ANS/0114.

2.2 Plant Extraction

The plant was open air-dried under shade, pulverized in a wooden mortar using pestle to

obtain a coarse material which was sieved and stored in plastic containers until used. 100 g of the powdered stem bark was soaked in 1000 mL of distilled water for 74 h, after which it was filtered using sterile white muslin cloth to remove debris. The solution again was filtered through a Whatman No.1 filter paper. The filtrate was then dried using a hot air oven at reduced temperature (40^{o}C). The dried extract was reconstituted in distilled water at 10 g/100mL. The reconstituted extract was labeled and kept in capped plastic containers at 4^{o}C until required.

2.3 Acute Toxicity Study

The method described by Lorke (1983) was used for the acute toxicity study. The study was conducted in two phases. In the first phase, three groups of four mice each were administered with the extract at respective oral doses of 10, 100 and 1000 mg per kg body weight. The animals were observed for signs of toxicity and possible deaths for 24h, 72h and 2 weeks. In the second phase, another three groups of four mice each were administered at respective oral doses of 1600, 2900 and 5000 mg per kg body weight of the extracts. They were equally monitored as in phase one for toxicity signs and deaths. From the data obtained, LD50 was determined.

2.4 Experimental Animals

Adult (male and female) albino rats weighing between 80-120 g were used for this research work. The animals were wire housed in aluminum cage under standard conditions. They were maintained on standard pellets and water *ad libitum*. The animals were acclimatized for 4 weeks before commencement of the experiment.

2.5 Grouping of Animals

The animals were grouped into four groups of five (5) albino rats each as follows:

Group A – Normal control (distilled water)
Group B – Diabetic rats untreated
Group C – Diabetic rats treated with 100mg/kg of aqueous extract of shea butter
Group D – Diabetic rats treated with 0.5mg/kg anti-diabetic drug (Glibenclamide).

2.6 Induction of Diabetes Mellitus

Stock solution of 5% alloxan was prepared by dissolving alloxan hydrate (1g) in 20 mL of distilled water. A day prior to the induction, the rats were allowed to fast overnight (6pm to 8am). Diabetes was induced by a single subcutaneous administration of alloxan hydrate (150 mg/kg body weight). The rats with blood glucose level greater than 150 mg/dl, two days post-induction (48-hour), were considered as diabetic and were treated with 1 mL of 100 mg/kg aqueous extract of the plant for two weeks. Their blood glucose levels were monitored after every four days using Fine test glucometer. Blood samples were collected from the tail of the rats after every four days to measure the glucose level.

2.7 Biochemical Assay

On the last day of treatment, the animals were anaesthetized with chloroform and 5 mL of blood sample was collected via cardiac puncture. The blood samples were collected into plain bottles and centrifuged at 5000rpm for 10 minutes to obtain serum. The serums obtained were used for the measurement of glucose, urea, creatinine and sodium levels. Serum glucose was estimated using glucose oxidase method (Trinder, 1954), urea was determined by urease method (Ambigas and Muthuraman, 2010), creatinine was assayed with the method described by Jaffe's (1841) while sodium was determined using Flame photometer.

2.8 Statistical analysis

The results obtained from the assays were expressed as mean ± SEM. Analysis of variance was used to test for differences in the groups using Instant Graphpad statistical software. Differences were considered to be statistically significant at $p< 0.05$.

3. Results and Discussion

Table 1. Record of Mortality in the Phase I Toxicity Study

Extract dose (mg/kg body weight)	Mortality
10	0/4
100	0/4
1000	0/4

Number of deaths in the group = 0
Number of mice in each group = 4

Table 2. Record of Mortality in the Phase II Toxicity Study

Extract dose (mg/kg body weight)	Mortality
1600	0/4
2900	0/4
5000	0/4

Number of deaths in the group = 0
Number of mice in each group = 4

From the results above, no mortality was observed in all the phases (Tables 1 & 2). No physical change such as, fur, raised tails, salivation, paw licking was observed which indicated that the stem bark extract did not cause any noticeable adverse effect in the animals. Base on the outcome of this study, Shea tree (*Vitellaria paradoxa)* may not cause acute toxicity

at a dose less than or equal to 5000 mg/kg in mice, which should be considered as practically harmless (OECD, 2001). This finding agrees with similar study of Fabricant and Farnsworth (2001) conducted, in which acute toxicity of methanol *Annona Senegalensis* leaf extract was evaluated in albino mice.

Blood glucose levels of the four groups of albino rats are presented in Table 3, which shows the blood glucose levels of the rats two days after induction of diabetes, 4th, 8th and 14th days of treatment with the extract. The blood glucose level of alloxan induced diabetic rats (Group B) significantly increases (p<0.05) from induction to the last day of experiment (Table 3). This confirmed the development of hyperglycemia. The diabetic untreated and treated rats showed symptoms of diabetes, such as polyphagia, polyuria and polydypsia before administration of the extract. The results of this study reveals that treatment of group C rats with aqueous stem bark extract of *Vitellaria paradoxa* (Shea butter) results in significant (p<0.05) lowering of blood glucose levels when compared with group B. This may be due to increase in peripheral glycolysis by increasing the activity and amount of several key enzymes (glucokinase, pyruvate kinase and phosphofructokinase). Enhanced glycolysis increase glucose utilization and thus indirectly decreases glucose release into plasma. Previous reports have indicated that plant extracts possess hypoglycemic properties, possible insulin release stimulatory effects and uptake of peripheral glucose, which in turn reversed alloxan induced hyperglycemia (Okonkwo and Okoye, 2009; Luka and Tijjani, 2013).

Table 3. Serum Glucose Levels of Alloxan-Induced Diabetic Rats Treated with Aqueous Stem Bark Extract of Shea tree (*Vitellaria Paradoxa)*.

Groups	2nd day after induction	4th day of treatment	8th day of treatment	14th day of treatment
A (Normal, control)	114± 4.68	105±2.89	95±2.89	109±0.58^{b}
B (Diabetic untreated	158±3.98	165±2.33	170±2.77	168±0.58^{a}
C (Diabetic treated with 100 mg/kg extract)	155±2.35	130±2.46	116±3.18	100±0.29^{b}
D (Diabetic treated with 0.5 mg/kg Glibenclamide)	160±5.67	125±2.69	111±5.56	96±5.777^{b}

Values are the Mean ± SEM (n=5). Mean values with different superscript letters are significantly different (p< 0.05)

Table 4. Serum Urea, Creatinine and Sodium Levels of Alloxan-Induced Diabetic Rats Treated with Stem Bark Extract of Shea Tree (*Vitellaria Paradoxa)*.

Groups	Concentration of Biochemicals (mg/dl)		
	Urea	Creatinine	Sodium
A (Normal, control)	35.5±7.42	338.5±69.00***	119.75±7.26^{c}
B (Diabetic untreated	110.25±33.03^{b}	737±126.53**	136.75±9.75^{b}
C (Diabetic treated with 100 mg/kg extract)	51.00±3.24^{c}	638±201.53*	133±23.35^{a}
D (Diabetic treated with 0.5 mg/kg Glibenclamide)	45.5±4.57^{c}	626±119.67*	125±1.78^{a}

Values are the Mean ± SEM (n=5). Mean values with different superscript letters and asterisk between the groups (or columns) are significantly different (p< 0.05)

There is significant (p< 0.05) decrease in the serum levels of urea, creatinine and sodium in diabetic rats treated with the extract when compared with diabetic untreated rats (Table 4). Whereas, no significant difference (p>0.05) was observed in serum concentrations of urea, creatinine and sodium in diabetes rats treated with the extract and those treated with the standard diabetic drug, indicating the ability of the extract to maintain the levels of these kidney function biomarkers in diabetic rats. Previous report has indicated that plant extract possesses the ability to significantly decrease serum urea and serum creatinine in diabetic rats treated with *M. emarginata (*Rajiv and Sasikumar, 2012).

4. Conclusion

This study has shown that aqueous stem bark extract of Shea butter tree *(Vitellaria paradoxa)* may be safe at doses less than or equal to 5000 mg/kg in mice, and normalizes blood glucose, urea, creatinine and sodium levels of alloxan induced diabetic rats. Thus, the findings suggest that the extract may be used as a useful screening material for antidiabetic drug.

Conflict of interest

The authors declare no conflict of interest.

References

Ayankunle, A. A., Kolawole,O.T., Adesokan, A.A. and Akiibinu, M.O. (2012). Antibacteria activity and sub-chronic Toxicity Studies of *vitelleria paradoxa* stem bark extract. *Journal of pharmacology and Toxicology*, *7*(6), 298-304.

Akinpelu, D. A. and Onakoya, Z. T. M. (2006). Antimicrobial Activities of Medicinal Plants used in Folklore Remedies in South-western Nigeria. *African Journal of Biotechnology*, 7(5), 1078-1081.

Alfred, T. (2002). "Fats and Fatty Oils". Ullmann's Encyclopedia of Industrial Chemistry. *Weinheim: Wiley-VCH.*

Aronson, D. and Rayfield, E.J. (2002). How Hyperglycemia Promotes Atherosclerosis: Molecular Mechanisms. *Cardiovascular Diabetology,*1(1). doi:10.1186/1475-2840-1-1

Fabricant, D. S. and Farnsworth, N. R. (2001). The Value of Plants used in Traditional Medicine for Drug Discovery. *Environmental Health perspectives*, *109* (1) 69-75.

Kumar, P. and Clark, M. (2002). *Diabetes Mellitus and Other Disorders of Metabolism* (2nd ed.). London, Elsevier, 106**9-**71.

Lorke, D. (1983). A New Approach to Tropical Acute Toxicity Testing. *Archives of Toxicology*, 54(4), 275-287.

Luka, C.D. and Tijjani, H. (2013). Comparative Studies of the Aqueous Extracts of Ocimum gratissimum, Aloe vera, Brassica oleracea and Ipomoea batatas on Some Biochemical Parameters in Diabetic Rats. *International Organization of Scientific Research Journal of Pharmacy and Biological Sciences*, *6*(3), 23-29.

Ndukwe I. G., (2007). Phytochemical screening of the crude extract of the roots, stem bark and leaves of *Vitallaria paradoxa* (GAERTN. F). *African Journal of Biotechnology, 6*(16)1905-1909.

Okonkwo, P.O. and Okoye, Z.S.C. (2009). Hypoglycaemic Effects of the Aqueous Extract of Newbouldia Leaves Root in Rats, *International Journal of Biological and Chemical Sciences, 3*(5), 998-1004.

OECD (2001). OECD Guidelines for testing chemical. Acute Oral toxicities, up and down procedure. 423, 1-14.

Trinder, P. (1954). Rapid determination of salicylate in biological fluids. *Biochemical Journal, 57* (2), 301–303.

Rajiv, G., G. and Sasikumar, P. (2012) Antidiabetic effect of *Merremia emarginata* Burm. F. in streptozotocin induced diabetic rats. *Asian Pacific Journal of Tropical Biomedicine*, *2*(4), 281–286.

Yakubu, M. T., Bilbis, L. S., Lawal, M. and Akanji, M. A. (2003). Effect of repeated administration of sildenafil citrate on selected enzyme activities of liver and kidney of male albino rat. *Nigerian Journal of Pure and Applied Sciences*,18(1),1395–4000.

