

# GSJ: Volume 5, Issue 11, November 2018, Online: ISSN 2320-9186 www.globalscientificjournal.com

# Antidiabetic and haematological effect of aqueous extract of Root of *spondias mombin* on streptozotocin-induced diabetic albino rats

<sup>1</sup>Nwaogwugwu, Joel Caleb\*, <sup>1</sup>Uhegbu, F.O, <sup>1</sup>Atasie, O.C., Ezekwe, A.S<sup>2</sup> and <sup>1</sup>Egege Amaka.

<sup>1</sup>Department of Biochemistry, Faculty of Biological Sciences, Abia State University, Uturu.

<sup>2</sup>Department of Medical Biochemistry, Imo State University, Owerri.

#### \*ifesinachinwa@gmail.com

## ABSTRACT

To investigate the antidiabetic properties of aqueous extract of root of spondias mombin and its beneficial effect on haematological parameters in streptozotocin induced diabetic rats. A total of 30 rats including 24 diabetic and 6 normal rats were used for this study. Diabetes was induced in male Wistar rats by intraperitoneal injection of streptozotocin. After being confirmed diabetic, animals were orally treated extracts at 200,400 and 600 mg/kg body weight daily for 14 days. The haematological parameters including red blood and white blood cells and their functional indices were evaluated in diabetic treated groups compared with the controls. The extract significantly reduced the blood glucose levels while the best result was obtained at 400 mg/kg body weight. The study also demonstrated improvement in the in vivo antioxidant property of Spondias mombin root extract showed marked increase in the levels of catalase and peroxidases and a decrease in the levels of Thiobarbituric acid reactive substances ( the malondialdehyde levels). The feed and water intake in diabetic rats were significantly reduced while weight loss was minimized at both dosages. Similarly, the levels of red blood, white blood cells and their functional indices were significantly improved after extract administration at both doses. It can be concluded that the aqueous extract of bark of spondias mombin possesses antihyperglycemic properties. In addition, the extract can prevent various complications of diabetes and improve some haematological parameters.

**Keywords:** *Spondias mombin,* Diabetes mellitus, Haematology, Antidiabetic property, Aqueous extract, Haematological parameter.

446

# 1. Introduction

Diabetes mellitus (or diabetes) is a chronic, lifelong condition that affects the body's ability to use the energy found in food due to absolute or relative deficiency of insulin secretion with/without varying degree of insulin resistance[1]. There are three major types of diabetes: type 1 diabetes, type 2 diabetes, and gestational diabetes. Diabetes mellitus is а chronic metabolic disorder of carbohydrates, proteins and fat. The number of people suffering from diabetes worldwide is increasing at an alarming rate[2].

The presently, diabetes can't be cured, but it can be managed and controlled. The goals of managing diabetes are to: Keep blood glucose levels as near to normal as possible by balancing food intake with medication and activity, blood cholesterol Maintain and triglyceride (lipid) levels as near their ranges as normal possible bv decreasing the total amount of fat to 30% or less of total daily calories and saturated bv reducing fat and cholesterol, and blood pressure control. Meanwhile none of the antidiabetic drugs could give a long term glycaemic control without causing any adverse side effects [3].

Therefore medicinal plants that are effective in controlling plasma glucose level with minimal side effects are commonly used in under developed countries as alternative therapy. In Africa, hundreds of plants are used traditionally for the management and/or control of diabetes mellitus. Unfortunately, only a few of such medicinal plants have been scientifically validated [4].

Spondias mombin is a tree, a species of flowering plant in the family anacardiaceae. It is commonly known as hog plum. It is a small deciduous tree that grows up to 20m high and 1.5m in girth, moderately buttressed; bark thick, corky, deeply fissured, slash pale pink, darkening rapidly and branches low. The fruits have a sharp, somewhat acidic taste and are edible. Their flesh surrounds a spiny kernel. It is native to the tropical Americas including the West Indies and has been naturalized in parts of Africa, India, Bangladesh, Sri Lanka and Indonesia. It has been widely credited with a lot of medicinal properties among which are its usage as a diuretic. The bark is used as a purgative, causing relief through vomiting. The leaves are effective for worms in children and has been applied as an eye lotion. The plant is also useful as an antidiarrhoeal agent, as an antimicrobial agent, and as an oxytocic and astringent [5],[6],[7],[8]. The extract has been documented to have antiinflammatory activity in Wistar rats. S. mombin leaf extract exerts antidiabetic properties. Therefore, this study is to aimed at assessing the antidiabetic properties and beneficial effects of aqueous extract of Spondias haematological mombin root on parameters in streptozotocin - induced diabetic rats.

# 2. Materials and methods

# 2.1. Plant materials

Fresh root of *Spondias mombin* was collected in Okigwe, Imo State, Nigeria in April 2018. The plant was identified and authenticated by Prof C.I Ogbonnaya of the Department of Botany, Abia State University, Uturu, Abia State Nigeria. The voucher sample was prepared and deposited in the herbarium of the Department Botany.

## 2.2. Preparation of the extract

The root material of Spondias mombin was sun-dried to constant weight in the laboratory. The dried material was then pulverized using an electric blender Products (Waring Division. Torrington, USA). About 40 g of the powdered plant material was extracted in 1 L of cold sterile distilled water maintained on a mechanical shaker (Stuart Scientific Orbital Shaker, UK) for 48 h. The extract was filtered using a Buchner funnel and Whatman No.1 filter paper. The filtrate was quickly frozen at -40 °C and dried for 48 h using a freeze drver (Savant Refrigerated vapor Trap, RV T41404, USA) to give a yield of 6.4 g of dry extract with brownish colour. The resulting extract was reconstituted with cold distilled water to give desired doses of 200 and 400 and 600 mg/kg body weight.

# 2.3. Animals

Assumed health male Wistar rats weighing between 150 and 180 g were obtained from the animal house of college of medicine of University of Nigeria Nsukka. They were kept in well ventilated house conditions [temperature (28±1) °C; photoperiod: 12 h light and 12 h dark cycle; humidity: 45%-50%]. The animals were allowed free access to food and water through out the investigation ad libitum. Ethics clearance was approved by the Animal Ethics Committee of Abia State University, Uturu.

# **2.4. Induction of diabetes in the rats**

Diabetes was induced in overnight fasted male Wistar rats by a single intraperitoneal injection (i.p.) of freshly prepared solution of streptozotocin (50 mg/kg body weight) in 0.1 M citrate buffer (pH 4.5). The animals were confirmed diabetic by the elevated plasma glucose levels after 72 h of injection. The rats with stable glycosuria and hyperglycemia (blood glucose >8.1 mmol/L) were used for the experiment[9].

# 2.5. Experimental design

Thirty male rats were randomly placed into five groups consisting of six animals in each group. Group I served as normal control rats and were allowed drinking water daily ; Group II served as negative control and were allowed to feed and water only; Group III : diabetic rats, treated daily with (glimepiride(0.6 Amarvl mg/kg). Group IV were administered 200 mg/kg body weight of Spondias mombin root extract, Group V were administered 400 mg/kg body weight of Spondias mombin root extract, VII were administered 600 mg/kg body weight of Spondias mombin root extract. The test animals were treated for 14 days through gavage tube orally.

# **2.6. Determination of haematological parameters**

The Horiba ABX 80 Diagnostics (ABX pentra Montpellier, France) was for the determination used of hematological parameters including red blood cells (RBC) and its related following indices manufacturer's instruction. These include hemoglobin (Hb), packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), corpuscular haemoglobin mean red concentration (MCHC). cell distribution width (RCDW). White neutrophils, blood cell (WBC), monocytes, lymphocytes, eosinophils, basophils and platelet were also analyzed[10,11,12].

# **2.7** Effect of extract on the weight, feed and water intake of the rats

Feed and water intakes were measured every day at the same hour during the experimental periods while the body weight of the animals were measured at zero day and every fifth day for the period of 14 days.

# 2.8 Lipid peroxidation assay (thiobarbituric acid reactive substances)

It was evaluated by thiobarbituric acid reactive substances (TBARS) tests an acid-heating reaction. during Aliquots of samples were incubated with 15% trichloroacetic acid and 0.38% thiobarbituric acid. The mixture was heated (1 h) in a boiling water bath. TBARS was determined by reading the absorbance of the pinkcomplex formed colored in a spectrophotometer at 532 nm[13].

# 2.9.1 Superoxide dismutase assay

This was estimated by the reaction mixture which contained 0.1 mL of phenazine methosulfate (186  $\mu$ L), 1.2 mL of sodium pyrophosphate buffer (0.052 mL; pH 7.0), 0.3 mL of the supernatant after centrifugation (1,500 × g for 10 min followed by 10,000 × g for 15 min) of homogenate was added to the reaction mixture. Enzyme reaction was initiated by adding 0.2 mL of NADH (780  $\mu$ M) and stopped after 1 min by adding 1 mL of glacial acetic acid. The amount of chromogen formed was measured by recording color intensity at 560 nm. Results were expressed in units/mg protein[14].

## 2.9.2 Catalase assay

It was determined with reaction solution contained 2.5 mL of 0.05 M phosphate buffers (pH 8.3), 0.7 mL of 0.2 M  $H_2O_2$  and 0.1 mL of tissue homogenate. Changes in absorbance of the reaction solution at 570 nm were determined after 1 min. Results were expressed in units/mg protein[15].

## 2.9.3 Reduced glutathione assay

This was estimated by using dithiobisnitro-benzoate as a substrate. The yellow color developed and read immediately at an absorbance of 412 nm and expressed as  $\mu$ M GSH/g protein[16].

# 2.9.4 Statistical analysis

Data were expressed as (mean  $\pm$  SD) of six replicates and were subjected to one way analysis of variance (ANOVA). Means were separated by the Duncan multiple test using SAS. Values were

#### 3. Results

 TABLE 1: Effects of Spondias mombin Aqueous Extracts root on Blood
 Blood

 Glucose Levels (mg/dl)
 Blood

Group	0	7	14	21	28
Normal control	$62.16 \pm 14.37^{a}$	58.83±6.11 <sup>a</sup>	61.33±.86 <sup>a</sup>	$67.33 \pm 14.58^{a}$	$60.83 \pm 0.65^{a}$
Diabetic control	$227.33 \pm 17.16^{\circ}$	$257.16 \pm 9.12^{d}$	$263.16 \pm 29.32^{d}$	$265.66 \pm 25.15^{d}$	$206.00 \pm 4.08^{\circ}$
Standard Drug(0.6mg/bw)	$233.50 \pm 28.11^{\circ}$	200.50± 2.53 <sup>c</sup>	$196.50 \pm 13.18^{b}$	$149.00 \pm 15.38^{a}$	$125.66 \pm 8.15^{d}$
200mg/kg	$224.33 \pm 19.81^{b}$	207.16± 4.72 <sup>b</sup>	$205.00 \pm 15.46^{b}$	198.16 ±11.53 <sup>b</sup>	165.50 ±04.78 <sup>a</sup>
400mg/kg	$229.83 \pm 24.49^{\circ}$	231.16± 7.13 <sup>c</sup>	$226.50 \pm 24.83^{\circ}$	$226.16 \pm 20.65^{\circ}$	161.00 ± 8.39 <sup>a</sup>
600 mg/kg	$236.33 \pm 22.10^{d}$	220.00± 6.74 <sup>c</sup>	$216.33 \pm 17.87^{b}$	$215.33 \pm 17.50^{\circ}$	$186.00 \pm 8.92$ <sup>c</sup>

Values are mean  $\pm$  SD for n=6. Values in the same row bearing the same letter of the alphabets are not significantly different (P >0.05) from each other.

The result of the effect of aqueous extract of *Spondias mombin* root on blood glucose level of streptozotocin – induced diabetic rats is presented in Table 1. The result revealed that the streptozotocin significant increased (P<0.05) the blood glucose level of the animals when compared to the normal control at days 7, 14, 21 and 28. Amaryl and plant extract slightly (P<0.05) at different concentration reversed the effect at different days. However, there was significant (P>0.05) difference in the blood glucose level of the animal between the normal and Diabetic control.

Parameters	Control group	Diabetes group	Amaryl (glimepiride) group	Extract at 200 Mg/Kg Bw	Extract at 400 mg/kg bw	Extract at 600 mg/kg bw
<b>RBC</b> (×10 <sup>12</sup> /L)	$\textbf{8.94} \pm \textbf{0.04}$	7.50 ± 0.60a	$\textbf{8.15} \pm \textbf{0.40b}$	$8.13 \pm 0.35^{b}$	$8.82 \pm 1.23^{b}$	$7.65 \pm 0.24$
Hb (g/dL)	$15.03 \pm 0.06$	13.43 ± 1.02a	$15.33 \pm 1.03b$	$15.10\pm0.84^{\rm b}$	$16.20 \pm 1.97^{\mathrm{b}}$	$12.03 \pm 0.05$
PCV (L/L)	$\textbf{0.50} \pm \textbf{0.02}$	0.36 ± 0.03a	$0.48 \pm 0.02 b$	$0.56 \pm 0.02^{b}$	$\textbf{0.60} \pm \textbf{0.07}^{b}$	$\textbf{4.70} \pm \textbf{0.08}$
MCV (fl)	55.83 ± 1.11	52.13 ± 0.38a	$62.65 \pm 1.02 b$	$68.30 \pm 2.52^{b}$	$68.50 \pm \mathbf{0.14^{b}}$	$50.33 \pm 0.10$
MCH (pg)	$16.93 \pm 0.35$	15.20 ± 0.40a	$17.80 \pm 0.46b$	$18.60\pm0.69^{b}$	$18.30\pm0.21^{\rm b}$	$20.50\pm0.32$
MCHC (g/dL)	$30.27 \pm 0.82$	18.23 ± 0.83a	$28.50 \pm 1.25 \mathrm{b}$	$27.30 \pm 0.66^{b}$	$26.80\pm0.35^{\mathrm{b}}$	$31.48 \pm 0.32$
RCDW (%)	13.63 ± 0.90	12.43 ± 0.55a	15.13 ± 1.20b	$14.50 \pm 3.13^{b}$	$13.50 \pm 2.47^{b}$	$10.98 \pm 0.10$

Table 2: The effect of aqueous extract of *Spondias monbin* root on red blood cells and the differentials in STZ induced diabetic rats (n = 6, mean  $\pm$  SD).

a: P < 0.05 vs control group; b: P < 0.05 vs diabetes group.

Table 3 shows the levels of serum WBC, basophils, neutrophils, eosinophils, lymphocyte and monocytes. The level of WBC was slightly increased after oral administration of the extract at 200 mg/kg while the dose of 400 and 600 mg/kg did not have any effect as compared with the diabetic groups. The plant extract significantly increased the level of lymphocyte, eosinophils, monocytes and platelet at both dosages while the best result was observed at the lower dose (200 mg/kg) (P<0.05). The significant effect depicted at 200 mg/kg compared favourably well whereas that of 200 mg/kg significantly boosted the level of neutrophils (Table 3). The extracts at dosages 200,400 and 600 did not have any beneficial effect on the level of basophils.

Parameters	Control	Diabetes group	Amaryl (glimepiride) group	Extract at 200mg/kg bw	Extract at 400mg/kg bw	Extract at 600 mg/kg bw
WBC (×10 <sup>9</sup> /L)	17.00 ± 3.20	$2.53\pm0.93^a$	$6.00\pm5.60^{b}$	$6.96 \pm 2.03^{b}$	$3.16 \pm 3.72^{b}$	30.32± 0.99
Neutrophils (%)	25.37 ± 1.17	$2.59\pm0.51^{a}$	$23.30 \pm 0.16^{b}$	$19.80\pm0.01^{\rm b}$	$40.60 \pm 0.30^{b}$	35.86 ± 0.05
Monocytes (%)	17.46 ± 6.11	$4.69 \pm 1.00^{a}$	$19.00 \pm 0.64^{b}$	$14.70\pm0.41^{\rm b}$	$9.60 \pm 0.01^{b}$	33.14 ±0.78
Lymphocyte (%)	65.40 ± 6.86	$5.36 \pm 0.33^{a}$	$61.40 \pm 5.10^{b}$	$60.90 \pm 2.20^{\mathrm{b}}$	$47.30 \pm 3.10^{b}$	29.41 ±0.16
Eosinophil (%)	5.70 ± 1.18	$1.03\pm0.78^{\rm a}$	$1.20 \pm 0.32^{b}$	$4.50 \pm 0.22^{b}$	$2.50 \pm 0.60^{b}$	38.75 ±0.37
Basophils (%)	0.53 ± 0.21	$0.03 \pm 0.03^{a}$	$0.25 \pm 0.15^{b}$	$0.10 \pm 0.02^{b}$	$0.10 \pm 0.03^{b}$	8.20±0.35
Platelets (×10 <sup>9</sup> )	851.00±78.58	55.00±31.11 <sup>a</sup>	176.00±55.20 <sup>b</sup>	201.00±68.00 <sup>b</sup>	90.00±20.00 <sup>b</sup>	7.91± 0.17

# Table 3: The effect of aqueous extract of *Spondias mombin root* on white blood cells and its differentials in STZ induced diabetic rats (n = 6, mean $\pm$ SD).

a: P < 0.05 vs control group; b: P < 0.05 vs diabetes group.

Group	CATALASE (IU/L)	PEROXIDASE	THIOBARBITURIC ACID REDUCING SUBSTANCE (TBARS) (µM/L) (MDA
		(I U/L)	activity)
Control	$0.0074 \pm 0.007$	$0.0081 \pm 0.009$	$0.181 \pm 0.108$
Diabetic group	$0.0010 \pm 0.001$	$0.0013 \pm 0.002$	$0.291 \pm 0.013$
Amaryl (glimepiride) group	$0.0076 \pm 0.003*$	$0.0096 \pm 0.003*$	0.125 ± 0.013 *
Extract at 200mg/kg bw	0.0061 ±0.001*	$0.0074 \pm 0.002$ *	0.148 ± 0.098 *
Extract at 400mg/kg bw	$0.0047 \pm 0.006*$	0.0056 ± 0.003 *	0.155 ± 0.054 *
Extract at 600mg/kg bw	$0.0052 \pm 0.004*$	$0.0054 \pm 0.003$ *	0.165 ± 0.083 *

**TABLE 4:** The effect of aqueous extract of *Spondias mombin root* on *invivo* antioxidant properties in STZ induced diabetic rats

Values are the mean  $\pm$  Standard deviation (SD) n=6, \*The mean difference is significant the p<0.05 level from the controls group.

There was significant increase in catalase (IU/L), peroxidase (IU/L) and significant decrease in thiobarbituric acid reducing substance (TBARS) ( $\mu$ M/L) (MDA Activity) in animals in group 3 (three) to 6 when compared to animals in group 2



Figure 1.: The effect of aqueous extract of *Spondias mombin* root on the feed intake of diabetic rats.

Values are mean  $\pm$  SD of 6 rats in each group. Normal: Control., Diabetic: without treatment., STD: Standard Drugs [Amaryl (glimepiride) ] D1 = Diabetic + Spondias mombin (200 mg/kg), D2 = Diabetic + Spondias mombin (400 mg/kg). D3 = Diabetic + Spondias mombin (600 mg/kg).



Figure 2.: The effect of aqueous extract of *Spondias mombin root* extract on the water intake of diabetic rats.

Values are mean ± SD of 6 rats in each group..Normal: Control., Diabetic: without treatment., STD: Standard Drugs [Amaryl (glimepiride) ] D1 = Diabetic + Spondias

# *mombin* (200 mg/kg), D2 = Diabetic + Spondioas mombin (400 mg/kg). D3 = Diabetic + Spondias mombin (600 mg/kg).

A significant decrease in the body weights (28-33 g) of diabetic animals was observed 10 days after induction of streptozotocin into the animals. The oral administration of plant extract markedly increased the body weight of the animals but the effect was not dose related (Figure 3). The percentage increase in the body weight at 400 mg/kg was 8.82% while that of 200 mg/kg did not show any significant difference as compared with the initial body weight.



Figure 3.: The effect of aqueous extract of *Spondias mombin root* extract on the Body Weight of diabetic rats. Values are mean ± SD of 6 rats in each group.

# **4** Discussion

Diabetes is induced by streptozotocin (STZ), a glucosamine–nitrosourea compound derived from *Streptomyces achromogenes* that is used clinically as a chemotherapeutic agent in the treatment of pancreatic  $\beta$  cell carcinoma. STZ damages pancreatic  $\beta$  cells, resulting in hypoinsulinemia and hyperglycemia[17,18]. STZ can induce a diabetic state in 2 ways, depending on the dose. The selectivity for  $\beta$  cells is associated with preferential accumulation of the chemical in  $\beta$  cells after entry through the GLUT2 glucose transporter receptor: chemical structural similarity with glucose allows STZ to bind to this receptor. The mode of action has best been demonstrated in mouse studies. At high doses, typically given singly, STZ targets  $\beta$  cells by its alkylating property corresponding to that of cytotoxic nitrosourea compounds[19]. At low doses, generally given in multiple exposures, STZ elicits an immune and inflammatory reaction, presumably related with the release of glutamic acid decarboxylase autoantigens. Under this condition, the destruction of  $\beta$  cells and induction of the hyperglycemic state is associated with inflammatory infiltrates including lymphocytes in the pancreatic islets[20]. STZ has well-known adverse side effects, which include hepatotoxicity and nephrotoxicity [21,22].

Streptozotocin is used as an agent to induce diabetes mellitus by selective cytotoxicity effect on pancreatic beta cells. Thus it affects endogenous insulin release and as a result increases blood glucose level. The continuous administration of aqueous extract of Spondias mombin aqueous root extract at 200.400 and 600 mg/kg or amarvl (glimepinde) for 14 days significantly reduced the blood glucose concentration in STZ induced diabetic rats. The plant extract (200,400 and 600 mg/kg) showed a comparable activity with the amaryl treated groups.

Amaryl is an oral blood sugar-lowering drug that is used to treat patients with type 2 diabetes. It contains the active ingredient glimepiride and belongs to the sulphonylurea class of diabetes medicines, which work by boosting the secretion of natural insulin and increasing the body's sensitivity to the blood sugar-regulating hormone. The probable mechanisms of action of the plant extract at higher dose could be linked to potentiation of insulin from beta cells or by increasing peripheral glucose uptake [21,22].

The assessment of haematological parameters could be used to reveal the

deleterious effect of foreign compounds including plant extracts on the blood constituents of animals. They are also used to determine possible alterations in the levels of biomolecules such as enzymes, metabolic products, haematology, normal functioning and histomorphology of the organs [23,24].

The occurrence of anaemia in diabetes mellitus has been reported due to the increased non-enzymatic glycosylation of RBC membrane proteins [15]. Oxidation of these proteins and hyperglycaemia in diabetes mellitus causes an increase in the production of lipid peroxides that lead to haemolysis of RBC [27]. In this study, the RBC membrane lipid peroxide levels in diabetic rats were not measured. However, the red blood cells parameters such as Hb, MCHC, MCH, PCV, MCV and RCDW were studied to investigate the effect of Spondias mombin root extract on the anaemic status of the diabetic rats. The levels of RBC, Hb, haematocrit, LUC and MCHC in the diabetic animals were drastically reduced which may be attributed to the infections on the normal body systems. This observation agrees with report al[20]who reported of Baskar et antihyperglycemic activity of aqueous root extract of Rubia cordifolia in streptozotocininduced diabetic rats. The alterations of these parameters are well known to cause anaemic condition in man [28]. Following plant extract administration, the level of RBC and its related indices were appreciably improved especially at 400 mg/kg. This gives an indication that the plant extract may contain some phytochemicals that can stimulate the formation or secretion of erythropoietin in the stem cells of the animals. Erythropoietin is a glycoprotein hormone which stimulates stem cells in the bone marrow to produce red blood cells [29]. The stimulation of this

hormone enhances rapid synthesis of RBC which is supported by the improved level of MCH and MCHC [30]. These parameters are used mathematically to define the concentration of haemoglobin and to suggest the restoration of oxygen carrying capacity of the blood. Though, the action mechanism of this plant is not investigated in this study. However, it may be attributed to the ability of plant extract to lower lipid peroxidation level that causes haemolysis of erythrocytes [31]. Previous study on this plant revealed presence flavonoids. the of proanthocyanidins, tannins, phenols and flavonols in this plant. These compounds have been reported to possess strong antioxidant capacity[32], therefore, could inhibit peroxidation of polyunsaturated fatty acids in the cell membrane and haemolysis of red blood cells in the diabetic animals reported by Torell and Faure et al [32]. Streptozotocin is a well known chemical that suppresses the immune system by damaging WBC and certain organs in the body [9]. intraperitoneal The injection of streptozotocin into rats significantly reduced the WBC count and its differentials such as basophils, monocytes, eosinophils, lymphocytes and neutrophils. The reduction of these parameters could be linked to suppression of leucocytosis from the bone marrow which may account for poor defensive mechanisms against infection [33]. Consequentially, they might have effects on the immune system and phagocytic activity of the animals [32]. The white blood counts and its related indices were significantly restored to near normal after plant extract administration at both doses. The presence of some phytochemicals with ability to stimulate the production of white blood count in the extract could be responsible for the observed result in the treated rats [34]. The extract at both dosages significantly improved the levels of WBC, monocytes, lymphocytes, eosinophils and

neutrophils as compared with glibenclamide treated group. However, the extract did not have any significant effect on basophils in this study.

Platelet aggregation ability has been shown in diabetic patient with long term poor glycaemic control due to lack or deficiency of insulin [35]. Platelets known as thrombocytes help to mediate blood clotting, which is a meshwork of fibrin fibres. The fibres adhere to any vascular opening and thus prevent further blood clot. It plays a crucial role in reducing blood loss and repairing of vascular injury[36]. The reduction of platelets levels in diabetic rats induced with streptozotocin was confirmed in this study in relation to the normal control rats. Long term reduction of this parameter may result in internal and external haemorrhage and finally leads to death. However, after plant extract administration, the level of platelet was improved markedly especially at the dose of 200 mg/kg while that of 400 mg/kg did not have strong effect as compared with diabetic untreated rats. This effect indicated the ability of the plant extract to stimulate the biosynthesis of clotting factors[23] due to the presence of active compounds that might help to precipitate blood coagulation or clotting, especially during severe bleeding or haemorrhage [32].

Also in this work there was increased levels of peroxidases, catalases and decrease level of thiobarbyturic acid reducing substances (levels of MDA) at p <0.05 when compared to the control groups. The increase in free radicals as described by (Kim *et al., [37]* is associated with oxidative processes. The feed and water intake of the diabetic rats were significantly increased as compared with the normal rats. These symptoms are well known markers of type 2 diabetes in both human and animal models which are direct consequence of insulin deficiency[1]. The feed intake was significantly reduced after administration of Spondias mombin root extract. The dose of 200 mg/kg showed higher activity than 400 mg/kg which was also reflected in water intake. The water intake of diabetic animals was significantly higher than the diabetic treated rats. The dose of 200 mg/kg was significantly lower than the group treated with 200 mg/kg. These results were similar to the report of Oyeluma et al., [19,37] who demonstrated the effect of *leonotis heorman* in controlling the desire for food and water intake under diabetic condition. A significant decrease in the body weights (28-33 g) of diabetic animals was observed 10 days after induction of diabetes in the animals. The loss in the body weight of the animals agrees with the finding of Oyedemi et al. [19] who observed similar effect on streptozotocininduced diabetic animals. This reduction has been linked to degradation of structural muscle proteins and wasting. Oral administration of plant extract at all doses was able to improve the body weight of the animals. The result indicated that extract of Spondias mombin root extract possessed the ability of managing glucose level as well as controlling muscle wasting and induced adipogenesis[21].

# Conclusion

From the data obtained in the present study we can conclude that the *Spondias mombin* root extract possesses antihyperglycemic properties. In addition, the extract could prevent various compications of diabetes as well as improving some haematological parameters. The study also demonstrated improvement in the *in vivo* antioxidant property of *Spondias mombin* root extract showed marked increase in the levels of catalase and peroxidases and a decrease in the levels of Thiobarbituric acid reactive substances (the malondialdehyde levels). Further experimental investigation is also needed to exploit its relevant therapeutic effect to substantiate its ethnomedicinal usage.

## REFERENCES

1. Mohammed A, Tanko Y, Okasha MA, Magaji RA, Yaro AH. Effects of aqueous leaves extract of *Ocimum gratissimum* on blood glucose levels of streptozotocininduced diabetic Wistar rats. *Afr J Biotechnol.* 2007;6(18):2087–2090.

2. Wild SG, Roglic A, Green R, Sicree R, King H. Global prevalence of diabetes: estimated for the year 2000 and projection for 2030. *Diabetes Care*. 2004;27(5):1047–1053.

3. Singh S, Loke YK, Furberg CD. Thiazolidinediones and heart failure: a teleoanalysis. *Diabetes Care*. 2007;30(8):2148– 2153.

4. Tanko Y, Yaro AH, Isa AI, Yerima M, Saleh MIA, Mohammed A. Toxicological and hypoglycemic studies on the leaves of *Cissampelos mucronata* (Menispermaceae) on blood glucose levels of streptozotocininduced diabetic Wistar rats. *J Med Plant Res.* 2007;2:113–116.

5. Abo K.A., Ogunleye V.O. and Ashidi J.S. (1999): Antimicrobial potential of *Spondias Mombin*, Croton zambesicus and zygotritonia crocea. *Phytotherapy Research.*, 13, pp 494-497

6. Iwu, M. M. (1993): Handbook of African Medicinal Plants, Boca Raton, p. 435. CRC Press

7. Kokwaro J.O. (1976): Medicinal Plants of East Africa. Nairobi, Kenya: East Africa Literature Bureau. Kampala. p. 384

8. Oliver-Bever, B. (1960): Medicinal plants in Nigeria. Being a Course of Four Lectures Delivered in April 1959 in the Pharmacy Department of the Nigerian College of Arts, Science and Technology, Ibadan. p. 760. University of Ibadan press.

9. Paul AJ. Streptozotocin. [Online] Available from: http://www.answers.com/topic/streptozotoci n#ixzz1KvPBTumc.

10. Jou JM, Lewis SM, Briggs C, Lee SH, De La Salle B, McFadden S. The International Council for Standardization in Haematology. An ICSH review of the measurement of the erythocyte sedimentation rate. *J Lab Hematol.* 2011;33(2):125–32

11. Horowitz GL, Altaie S, Boyd JC, Ceriotti F, Garg U, Horn P. The Clinical and Laboratory Standards Institute and IFCC. Defining, establishing, and verifying the reference intervals in a clinical laboratory; Approved Guideline-Third edition. C28-A3, Vol.28. No.30. 2012 June 20].

12. Demirin H, Ozhan H, Ucgun T, Celer A, Bulur S, Cil H.The normal range of the mean platelet volume in healthy subjects: Insights from a large epidemiologic study. *Thromb Res.* 2011;128(4):358–60.

13. Satoh K. Serum lipid peroxide in cerebrovascular disorders determined by a new colorimetric method. *Clin Chim Acta*. 1978;90:37–43.

14. Kakkar P, Das B, Viswanathan PN. A moditometric assay of superoxide dismutase. *Indian J Biochem Biophys*. 1984;21:131–2

15. Maehly AC, Chance B. I. New York: Interscience; 1954. Methods of Biochemical Analysis; pp. 357–8.

16. Ellman GL. Tissue sulfhydryl groups. *Arch Biochem Biophys.* 1959;82:70–7.

17. Swantson-Flatt SK, Day C, Bailey CJ, Flatt PR. Traditional treatments for diabetes: studies in normal and streptozotocin diabetic mice. *Diabetologia*. 1990;33:462–464.

18. Viswanathan M, Janarthanan VV, Rajendra P. The epidemiology of cardiovascular diseases in type 2 diabetes: The Indian Scenario. *J Diabetes Sci Technol.* 2010;4(1):158–70.

19. Oyedemi SO, Yakubu MT, Afolayan AJ. Antidiabetic activities of aqueous leaves extract of *Leonotis leonurus* in streptozotocin induced diabetic rats. *J Med Plant Res.* 2011a;5(1):119–125.

20. Baskar R, Bhakshu LM, Bharathi GV, Reddy SS, Karuna R, Reddy GK, Antihyperglycemic activity of aqueous root extract of *Rubia cordifolia* in streptozotocininduced diabetic rats. *Pharm Biol.* 2006;44(6):475–479.

21. Shenoy AG, Ramesh KG. Improvement of insulin sensitivity by perindopril in spontaneously hypertensive and streptozotocin induced diabetic rats. *Indian J Pharmacol.* 2002;34:156–164.

22. Oyedemi SO, Yakubu MT, Afolayan AJ. Antidiabetic activities of aqueous leaves extract of *Leonotis leonurus* in streptozotocin induced diabetic rats. *J Med Plant Res.* 2011;5(1):119–125.

23. Adebayo OJ, Adesokan AA, Olatunji LA, Buoro DO, Soladoye AO. Effect of ethanolic extract of *Bougainvillea spectabilis* leaves on haematological and serum lipid variables in rats. *Biokemistri*. 2005;17:45–50.

24. Oustamanolakis P, Koutroubakis IE, Kouroumalis EA. Diagnosing anemia in inflammatory bowel disease: beyond the established markers. *Crohns Colitis.* 2011;5(5):381–91

25. Balasubraimanian T, Lal MS, Mahananda S, Chatterjee TK. Antihyperglycaemia and antioxidant activities of medicinal plant *Stereospermum suaveolens* in streptozotocin-induced diabetic rats. *J Diet Suppl.* 2009;6(3):227–251.

26. Magalhaes P, Appell H, Duarte J. Involvement of advanced glycation end products in the pathogenesis of diabetes complication: the protective role of regular physical activity. *Eur Rev Aging Phys Act*. 2008;5(1):17–29.

27. Nastaran JS. Antihyperglycaemia and antilipidaemic effect of *Ziziphus vulgaris* L on streptozotocin induced diabetic adult male Wistar rats. *Physiol Pharmacol.* 2011;47(1):219–223.

28. Oyedemi SO, Bradley G, Afolayan AJ. Toxicological effects of the aqueous extract of *Strychnos henningsii* Gilg in Wistar rats. *J Nat Pharm*. 2010c;1:1.

29. Ohlsson A, Aher SM. Early erythropoietin for preventing red blood cell transfusion in preterm and/or low birth weight infants. *Cochrane Database Syst Rev.* 2006;3:CD004863

30. Abu-Zaiton AS. Antidiabetic activity of *Ferula asafoetida* extract in normal and alloxan induced diabetic rats. *Pak J Biol Sci*. 2010;13(2):97–100.

31. Ashafa AOT, Yakubu MT, Grierson DS, Afolayan AJ. Toxicological evaluation of the aqueous extract of *Felicia muricata* Thunb. leaves in Wistar rats. *Afr J Biotechnol.* 2009;6(4):949–954.

32. Torell J, Cillard J, Cillard P. Antioxidant activity of flavonoids and reactivity with peroxy radical. Phytochemistry. 1986;25(2):383–385.

33. Dahlback B. Advances in understanding pathogenic mechanisms of thrombophilic disorders. *Blood*. 2008;112:19–27.

34. Akinpelu DA, Aiyegoro OA, Okoh AI. *In vitro* antimicrobial and phytochemical properties of crude extract of stem bark of Afzelia africana (Smith) *Afr J Biotechnol*. 2008;7(20):3665–3670.

35. Jarald E, Joshi SB, Jain DC. Diabetes and herbal medicines. *Iran J Pharmacol Ther*. 2008;7(1):97–106.

36. Kumar Abbas, Fausto Aster. Robbins and Cotran Pathologic Basis of Disease. 8th edition. Elsevier; 2010.

37. Kim HK, Kim MJ, Cho HY, Kim EK, Shin DH. Antioxidant and antidiabetic effects of amaranth (*Amaranthus esculantus*) in streptozotocin-induced diabetic rats. *Cell Biochem Funct*. 2006;24:195–199.

# JSJ