

BIOCHEMICAL AND HEAMATOLOGICAL EVALUATION OF METHANOL STEM BARK EXTRACT OF *BOMBAX BUONOPOZENSE* ADMINISTERED TO STREPTOZOTOCIN- INDUCED DIABETIC RATS

^{*1}Akhigbemen, A. M. and ^{2,3} Idomeh, F. A.

¹Department of Pharmacology and Toxicology, Faculty of Pharmacy, University of Benin, Benin City 300001, Nigeria.

² Department of Chemical Pathology, University of Benin Teaching, Benin City Nigeria
 ³ Present Address; Department of Biochemistry, Basildon University Hospital, Mid and South Essex, Basilon, Essex, United Kingdom.

*Corresponding Author: E-mail: <u>abigail.omo-isibor@uniben.edu</u>, +2347032470846 Received: 28th Jan., 2024 Accepted: 14th Mar., 2024 Published: 1st June, 2024

ABSTRACT

Background: *Bombax buonopozense* is a tree native to the rainforests of the West African region. Locally, various parts of the plant have been prepared and used in treating diseases like; rheumatism, cough, dysentery, malaria, and other ailments.

Aim: This study was aimed at evaluating the chemical constituents of *B. buonopozense* as well as its toxicological effect in diabetic rats.

Methodology: Hyperglycaemia was induced in rats by intraperitoneal (i.p) injections of streptozotocin (STZ) at a dose of 45mg/kg body weight. Methanol stem bark extract of *B. buonopozense* was administered orally at increasing doses of 200, 400, and 800mg/kg body weight to the streptozotocin-induced hyperglycaemic rats for 14 days after which animals were sacrificed and hematological, biochemical marker were evaluated. The constituents of *Bombax buonopozense* were identified using gas chromatography-mass spectrometry (GC-MS).

Results: GC-MS analysis revealed the presence of 35 compounds corresponding to 35 different peaks. There were no significant effects on, hematological parameters, serum proteins, bilirubin, alanine transaminase, aspartate transaminase, alkaline phosphatase and lipids. Histologically the liver, kidney, heart, spleen and brain of the animals were essentially normal except in the pancreas where mild congestion was observed.

Conclusion: This study shows several compounds that could be responsible for the pharmacological effect of the plant. Furthermore, the plant is relatively safe when used in the treatment of diabetes mellitus.

Keywords: Diabetes mellitus, *Bombax buonopozense*, Chromatography-mass spectrometry Toxicity.

INTRODUCTION

Diabetes is a chronic, metabolic disease characterized by elevated levels of blood glucose (or blood sugar). For the past three decades, despite the significant progress made in the treatment of diabetes, the results of treatment in patients is still far from perfect. These treatments have some disadvantages, including drug resistance (reduction of efficiency), side effects, and even toxicity. For example, sulfonylureas lose their effectiveness after 6 years of treatment in 44% of patients (Kooti et al., 2016). A good number of plants contain carotenoids. flavonoids, terpenoids. alkaloids, glycosides and can often have anti-diabetic effects (Afrisham et al 2015).Danso *et al.* (2019) noted that Bombax buonopozense is one of the underutilized plants in West Africa due to its medicinal and nutritional purposes.

72

Citation: Akhigbemen, A. M. and Idomeh, F. A. (2024): Biochemical and Heamatological Evaluation of Methanol Stem Bark Extract of *Bombax buonopozense* Administered To Streptozotocin- Induced Diabetic Rats. BJMLS 9(1): 72 - 86

Bombax buonopozense also known as Red silk cotton plant or Gold Coast Bombax is perennial plant mainly found in rainy forest zones of West African countries like Sierra Leone, East Gabon and some part of Nigeria (Beentie and Smith, 2001). The most utilized part of this plant is the leaves, bark, root, stem and trunk. A greater population uses these plant parts for several medicinal purposes ranging from the treatment of swellings, fever. and convulsion to psychosis insanity. and Though no compounds have been identified in or isolated from the stem bark of Bonbax buonopozense. Chisom *et al.* (2014)revealed that several parts of this plant possess substantial quantity of nutrients such carbohydrates, proteins calcium. as magnesium, zinc as well as anti-nutrients such as oxalates, phytates and cyanide in minute concentrations. It therefore becomes identify compounds imperative to responsible for the bioactive nature of this plant in diabetic rats as well as evaluate the safety of this plant.

MATERIALS AND METHODS Experimental Animals

Male albino Wistar rats weighing (200 - 350)g) were obtained from the Animal House of Department of Pharmacology and Toxicology, Faculty of Pharmacy, University of Benin, Benin City, Nigeria. A total of number of 60 rats were used for the study which were kept in kept in plastic cages and housed at room temperature (24°C) and Humidity. They were allowed free access to dry rodent pellet feeds (Top Feeds Limited, Ibadan, Nigeria) and water (borehole). The bedding materials (wood shavings) of the cages were changed daily. experiments were carried All out in accordance with the National Institute of Health Guidelines for the Care and Use of laboratory Animals (NIH Publications No. 80-23) Revised in 2002.

Collection and extraction of Plant Materials

Dried stem barks of *Bombax buonopozense* were collected, identified and authenticated at the National institute for pharmaceutical research and development (NIPRD) Abuja, Nigeria. The voucher specimens were deposited in the herbarium for future reference.

The stem bark of *Bombax buonopuzense* was air-dried under a shade to a constant weight and milled to fine powder using a mechanical grinder. A 500 g of the powdered pulp was added to 2.5L of methanol and allowed to stand for 72 hours. The mixture was filtered using a clean piece of cloth and cotton wool. The filtrate was concentrated to dryness under reduced temperature and pressure in an oven. The dried methanol stem bark extract (MSBEx-BB) was stored at 4 °C until use.

Inducement of Diabetes Mellitus

The animals were fasted overnight and diabetes mellitus was induced by a single dose intraperitoneal injection of streptozotocin (45 mg/kg body weight) dissolved in freshly prepared 0.1M citrate buffer, pH 4.5. After administration the animals were allowed free access to feed and drink. After 48 h, the animals were tested for diabetes using the Accu-Check® Active Glucometer (Roche, USA) and any animal with blood sugar level > 200 mg/dl was considered diabetic (Szkudelski, 2001).

Experimental Design

The animals were selected into six groups of 10 rats each and treated orally for 1 week as follows;

Group 1:Diabetic animals treated withglibenclamide5mg/kg body weight dailyGroup 2:Diabetic animals treated with

MSBEx-BB 200mg/kg body weight daily Group 3: Diabetic animals treated with

MSBEx-BB 400mg/kg body weight daily

Group 4: Diabetic animals treated with MSBEx-BB 800mg/kg body weight daily

Group 5: Diabetic animals given 0.2ml distilled water daily

Group 6: Non-diabetic animals given 0.2ml distilled water daily.

Gas Chromatography-Mass Spectrometry (GC-MS) Analysis

Approximately (0.1 g) of the extract was dissolved in 10 mL of 70 % methanol. The mixture was allowed to stand for 1 to 2 h in a sealed test tube. The mixture was decanted, centrifuged and filtered using a micron filter into a 5 mL sample bottle. Analysis of the methanol extract was done using a gas chromatography instrument (Model-7890A, Agilent USA) (Olivia et al., 2021). The compounds were identified by name, molecular formula, and molecular weight by comparing the mass spectra obtained with those of standard spectra from National Institute of **Standards** & Technology (NIST) library.

Evaluation of Hematological Indices

samples collected Blood into ethylenediamine tetra acetic acid (EDTA) bottles were used for the evaluation of red blood cell count (RBC), packed cell volume (PCV), hemoglobin concentration (Hb), platelet count (PLT), mean cell volume (MCV), mean corpuscular hemoglobin concentration (MCHC), and total white blood cell counts (WBC) as well as their differentials (granulocytes, lymphocytes and monocytes). The samples were analyzed using an automated hematology system Junior Hematology (Diatron Abacus Analyzer, China) (Akhigbemen et al., 2018). **Biochemical Assays**

Blood samples collected in plain bottles were allowed to clot at room temperature for approximately 4 hr before centrifugation using a Hettich® centrifuge (Rototix 32A, Germany) at 4000 rpm for 10 min. The obtained sera were used for evaluation of biochemical parameters including alanine transaminase (ALT), aspartate transaminase (AST), alkaline phosphatase (ALP) and lipid profile using standard diagnostic test kits (Randox laboratories, UK) on Automated System (VIS-7220G, Clinical Biotech Engineering Management Company Limited, UK; Analyzer ISE 4000 SFRI, following the manufacturer's France) instructions. Total bilirubin (TB) and direct bilirubin (DB) determined was using Jendrassik-Grof method (Spencer and Price, Bayero Journal of Medical Laboratory Science, BJMLS

1977). Urea was assayed using modified diacetyl monooxime method (Marsh *et al.*, 1965), while creatinine was determined by the Jaffe's method (Chawla, 1999). Ion selective electrode machine was used for the assay of sodium, potassium, chloride and bicarbonate using the method described by (Burnett *et al.*, 2000).

RESULTS

GC-MS chromatographic evaluation of the extract

The phytochemical constituents of the MSBEx-BB with their specific retention time and peak wavelength of absorption for each compound identified is shown in Figure 1. The 35 compounds identified as well as their molecular formulas and weights are given in Table 1.

Effect of MSBEx-BB on hematological parameters

The extract did not significantly alter red and white blood cell parameters and its differential's when compared with values from control rats (Table 2 and 3 respectively).

Effect of MSBEx-BB on plasma urea, creatinine and electrolytes

Table 4 shows that 14 days treatment with the extract had no effect on values for Creatinine, urea, Na+, K+, Cl⁻ as well as bicarbonate ions when compared across the groups.

Effect on plasma lipids

There were no significant differences in the lipid parameters: total cholesterol (TC), high density lipoproteins (HDL), low density lipoproteins (LDL) and triglycerides when compared with the control (Table 5).

Effect on plasma proteins and bilirubin

Table 6 shows that there were no significant differences in the plasma concentration of proteins and bilirubin in extract-treated rats when compared with the control.

Effect on plasma enzymes

There were no significant changes in the plasma levels of alanine transaminase (ALT), aspartate transaminase (AST) and alkaline phosphatase (ALP) in (Figures 2 to 4).

74

Effects on the histology of selected organs In the liver (Figure 5), there were no major histological changes. The photomicrographs of the kidney are shown in Figure 6. There were no changes in the corpusules, interstitial space and tubules of extracttreated rats when compared to the control. In the heart, all vasculature were normal except mildly enlarged coronary artery seen animals that in animals treated with glibenclamide, as well as 400 and 800 mg/kg of the extract. Photomicrograph of the spleen showed the presence of lymphoid follicles and lymphocytes.

```
File :C:\msdchem\l\AROMATIC SCAN\DR ADEYINKA1.D
Operator : DR EME KA
Acquired : 6 Jul 2021 2:22 using AcqMethod AROMATIC SCAN 2.M
Instrument : ILORIN MSD
Sample Name: SAMPLE A
Misc Info :
Vial Number: 12
```



S/N	Name of compound	Retention time	Molecular weight(g/mol)	Molecular Formular
1	L-Methionine	2.418	149.21	C ₅ H ₁₁ NO ₂ S
2	Nickel, bis(1,1,1,5,5,5-hexafluoro -4-thioxo-2-pentanonato-O,S)-	2.581	504.9	$C_{10}H_{2}F_{12}NiO_{2}S_{2} \\$
3	2-Decanone	2.644	156.2652	$C_{10}H_{20}O$
4	Methacrylic acid, 2,3,4,6-tetrachlorophenyl ester	2.737	299.965	$C_{10}H_6C_{14}O_2$
5	l-Valine, N-capryloyl-, methyl ester	2.787	257.3691	C14H27NO3
6	Alpha-l-rhamnopyranose	3.532	164.16	$C_6H_{12}O_5$
7	Diethanolamine	3.720	105.1356	C4H11NO
8	2-Hexene, 5-methyl-, (E)-	3.851	98.1861	C7H14
9	Ethane, diazo-	4.045	56.07	
				$C_2H_4N_2$
10	1-Octanol, 3,7-dimethyl-	4.620	158.2811	$C_{10}H_{22}O$
11	2,7-Dimethyl-1,7-octadien-3-amine	5.377	153.26	$C_{10}H_{19}N$
12	1-Butanol, 3-methyl-, acetate	5.552	130.1849	$C_7H_{14}O_2$
13	Urethane	5.721	89.0932	C ₃ H ₇ NO ₂
14	Propanenitrile, 2-hydroxy-	5.996	71.0779	C ₃ H ₅ NO
15	Pentane, 1-(1-ethoxyethoxy)-	6.741	160.2539	C9H20O2
16	Hexanoic acid, ethyl Ester	7.222	144.2114	$C_8H_{16}O_2$
17	Silane, triethyl-	8.273	116.2767	$C_6H_{16}Si$
18	Indene	8.580	116.1598	C9H8
19 20	Undecane, 4,6-dimethyl Octanoic acid, ethyl ester	9.418 9.837	184.3614 172.2646	$\begin{array}{c} C_{13}H_{28} \\ C_{10}H_{20}O_2 \end{array}$
21	Fumaric acid, heptyl 2-methylcyclohex-1-enylmethyl ester	10.513	308.4	$C_{18}H_{28}O_4$
22	Methoxyacetic acid, tetradecyl ester	10.801	286.4	C17H34O3
23	Decanoic acid, ethyl ester	11.601	200.3178	$C_{12}H_{24}O_2$
24	9-Octadecenoic acid,	13.878	282.4614	$C_{18}H_{34}O_2$
25	Squalene	14.066	410.7180	C30H50
26	Bis(2-ethylhexyl) phthalate	14.228	390.5561	$C_{24}H_{38}O_4$
27	Phthalic acid, hexyl neopentyl ester	14.904	332.44	C ₂₀ H ₂₈ O
28	Pentadecanoic acid, 14-methyl-, methyl ester	15.079	270.4507	$C_{17}H_{34}O_2$
29	Dibutyl phthalate	15.567	278.3435	C16H22O4
30	Acetamide, 2,2,2-trifluoro-N-methy	15.955	199.25	C ₆ H ₁₂ F ₃ NOSi
31	Pyrimidine, 5-fluoro-2-dimethylamino-	16.174	141.15	C ₆ H ₈ FN ₃
32	9,12-Octadecadienoic acid (Z,Z)-,methyl ester	16.437	294.4721	$C_{19}H_{34}O_2$
33	Phytol	16.731	296.5310	$C_{20}H_{40}O$
34	l-Valine N-canryloyl- methylester	17 556	257 3691	$C_{14}H_{27}NO_{2}$
34	Propanamide, 3-bromo-	18.013	151.990	C ₃ H ₆ BrNO
35	Succinic acid, di(tetradec-11-enyl) ester	18,376	506.8005	$C_{32}H_{58}O_4$

Akhigbemen and Idomeh (2024) BJMLS, 9(1): 72 - 86 Table 1: GC-MS identified constituents of Methanol stem bark extract of *B. buonopozense*

Table 2: Effect of 14-day of	oral treatment w	vith methanol stem	bark extract of	bombax buonopozense	administered	to streptozotocin-	induced
diabetic rats on red blood p	parameters and	platelet count					

Dose	RBC	HCT(%)	MCV(fl)	MCHC(g/dl)	Hgb(g/dl)	PLT
	(x 106/µl)					(x 103/µl)
Diabetic animals + Glibenclamide 5mg/kg	6.71±0.12	36.00±1.00	53.30±0.57	40.80±0.25	14.60±046	607.30±133.7
Diabetic animals + BB 200mg/kg	7.65 ± 0.26	45.63±3.82	59.47±3.24	34.87 ± 2.60	15.73±0.27	486.30±42.47
Diabetic animals +BB 400mg/kg	7.50 ± 0.06	44.83±3.60	59.93±4.25	35.03±3.18	15.57±0.33	480.3±90.33
Diabetic animals +BB 800mg/kg	7.55 ± 0.39	42.47±1.52	56.53±1.25	37.77±3.07	16.13±1.59	520.00±107.7
Untreated Diabetic animals	7.27 ± 0.27	43.80±2.15	60.30 ± 0.815	32.33±0.39	14.20 ± 0.8	316.00±67.14
Non-diabetic animal	7.12 ± 0.21	45.30±2.25	63.6±1.29	31.73±0.08	14.40 ± 0.75	496.70±95.79

Data represents mean ± SEM; n=6 rats. Hgb, hemoglobin; RBC, red blood cells; PLT, platelets; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; HCT, hematocrit.

 Table 3: Effect of 14-day oral treatment with methanol stem bark extract of bombax buonopozense administered to streptozotocin- induced diabetic rats on white blood cells and the differentials

Dose	WBC (x 10 ³ /ul)	LYM (x 10 ³ /ul)	MO (x10 ³ /ul)	GR (x10 ³ /µl)
Akhiahaman and Idomah (2024).	$\frac{(\mathbf{X} \mathbf{I} 0 / \boldsymbol{\mu} \mathbf{I})}{16.77 + 2.09}$	$(x 10 / \mu f)$	$\frac{(\mathbf{XI0} \boldsymbol{\mu} \mathbf{I})}{12.07 \boldsymbol{\mu} 2.922}$	$\frac{(x10 / \mu)}{6.40 + 2.00}$
Akingbernen und Tuomen (2024).	10.77 ± 3.08	80.33±0.330	13.2/±3.832	0.40±2.90
Akhigbemen and Idomeh (2024):	7.933 ± 2.78	91.87±0.953	6.067±1.017	2.06±0.15
Akhigbemen and Idomeh (2024):	5.733±2.619	90.30±4.33	6.200±3.13	3.50±1.21
Akhigbemen and Idomeh (2024):	10.70 ± 2.001	89.37±3.12	7.800 ± 2.30	2.83±0.93
Akhigbemen and Idomeh (2024):	10.23 ± 2.03	92.97±2.171	4.044 ± 0.78	$3.00{\pm}1.50$
Akhigbemen and Idomeh (2024):	2.93±0.32**	94.23±0.62	$2.733{\pm}0.18^{*}$	3.03±0.62

*P<0.05; **P<0.01 when compared to positive control. Data represents mean \pm SEM; n=6 rats. WBC, white blood cells; GR, granulocytes count; LYM, lymphocyte; MO, monocyte.

77

Akhigbemen and Idomeh (2024) BJMLS, 9(1): 72 - 86

Table 4: Effect of 14-day oral treatment with methanol stem bark extract of bombax buonopozense administered to streptozotocin- induced diabetic rats on plasma urea, creatinine and electrolytes.

Dose	Cr (g/dl)	Urea (g/dl	Na+ (mmol/L)	K+ (mmol/L)	Cl- (mmol/L)	HCO3- (mmol/L)
Diabetic animals + Glibenclamide 5mg/kg	1.66±0.12	82.33±13.96	132.70±0.91	6.25±0.23	98.00±0.86	$15.83{\pm}1.01$
Diabetic animals + BB 200mg/kg	2.00 ± 0.85	83.67 ± 0.38	137.00±3.60	5.77 ± 0.55	101.3 ± 2.67	17.67 ± 1.86
Diabetic animals +BB 400mg/kg	1.70 ± 0.60	66.00 ± 23.07	135.3±2.19	5.43 ± 0.32	102.3 ± 1.45	18.67 ± 1.20
Diabetic animals +BB 800mg/kg	1.63 ± 0.28	$64.67 {\pm} 10.91$	133.0±0.577	6.03±0.17	103.0±0.577	17.00 ± 1.53
Untreated Diabetic animals Non-diabetic animal	1.47±0.18 1.60±0.06	60.00±6.92 59.00±4.583	137.3±1.20 137.7±1.20	4.96±0.21 4.77±0.088	`103.3±1.45 `103.3±1.45	21.67±0.88 20.00±0.57

Values are not significantly different. Data represents mean \pm SEM; n=6 Cr, creatinine; Na⁺, sodium; K⁺, potassium; Cl⁻, chloride; HCO3⁻, bicarbonate.

Table 5: Effect of 14-d	ay oral treatment	with methanol st	em bark extrac	t of bombax	buonopozense	administered	to streptozotocin-	induced
diabetic rats on plasma	lipids of rats							

Dose	ТС	HDL	TG	LDL
	(mg/dl)	(mg/dl)	(mg/dl)	(mg/dl)
Diabetic animals + Glibenclamide 5mg/kg	127.00±11.82	36.00±2.61	51.50±7.293	80.50±13.3
Diabetic animals + BB 200mg/kg	152.00±19.08	40.00±4.933	60.00 ± 5.774	100.0±18.93
Diabetic animals +BB 400mg/kg	156.7±19.01	41.67±4.372	51.33±7.54	104.7±13.38
Diabetic animals +BB 800mg/kg	139.0±4.16	43.33±5.24	46.33±9.351	86.33±2.73
Untreated Diabetic animals	158.3±4.41	46.67±2.186	66.67±10.93	98.33±4.41
Non-diabetic animal	142.0 ± 9.07	41.33±1.33	60.00±7.635	88.67±9.493

Values are not significantly different. Data are represented as Mean \pm SEM; n=6 rats. TC, Total cholesterol; HDL, High density Lipoprotein; TG, Triglyceride; LDL, Low density lipoprotein.

78

Table 6:	Effect of 14-day oral	l treatment with	methanol sten	n bark extrac	t of bombax	buonopozense	administered t	o streptozotocin-	induced diabetic
rats on pla	asma protein and bilin	rubin of rats							

Dose	ТВ	ALB	ТР	GB
	(mg/dl)	(mg/dl)	(mg/dl)	(mg/dl)
Diabetic animals + Glibenclamide 5mg/kg	0.233 ± 0.02	3.86±0.07	7.00±0.07	3.133±0.03
Diabetic animals + BB 200mg/kg	0.40 ± 0.05	3.633±0.15	6.97±0.12	3.33±0.26
Diabetic animals +BB 400mg/kg	0.40 ± 0.10	3.56±0.03	7.20±0.20	3.63±0.21
Diabetic animals +BB 800mg/kg	0.40 ± 0.05	3.93±0.33	7.00±0.10	3.07±0.24
Untreated Diabetic animals	0.60 ± 0.05	3.80±0.11	7.00 ± 0.05	3.20±0.17
Non-diabetic animal	$0.77 \pm 0.15^{***}$	3.63 ± 0.08	6.93±0.15	3.30±0.21

****P<0.001 when compared to positive control. Data are presented as mean± SEM; n=6 rats. TB, Total Bilirubin; ALB, Albumin; TP, Total protein; GB, Globulin.



Figure 2: Effect of methanol stem bark extract of bombax buonopozense on alkaline phosphatase (ALP) following 14-day administration to streptozotocin-induced diabetic rats. Values are not significantly different from the control. All values are expressed as mean \pm SEM, n=6.



Figure 3: Effect of methanol stem bark extract of bombax buonopozense on alanine transaminase (ALT) following 14-day administration to streptozotocin- induced diabetic rats. Values are not significantly different from the control. All values are expressed as mean \pm SEM, n=6.



Figure 4: Effect of methanol stem bark extract of bombax buonopozense on aspartate transaminase (AST) following 14-day administration to streptozotocin- induced diabetic rats. Values are not significantly different from the control. All values are expressed as mean \pm SEM, n=6.



Figure 5: Representative photomicrographs of the liver of streptozotocin induced diabetic rats. A: positive control 5mg/kg glibenclamide, B: extract 200mg/kg, C: extract 400mg/kg, D: 800mg/kg, E:diabetic untreated, F:non-diabetic animals. Long arrow shows visible centriole while short arrows show hepatocytes as well as pyknotic nucleus. H & E, × 400



Figure 6: Representative photomicrographs of the kidney of streptozotocin induced diabetic rats. A: positive control 5mg/kg glibenclamide, B: extract 200mg/kg, C: extract 400mg/kg, D: 800mg/kg. Kidney reveals visible renal corpuscle (long arrow), interstitial space and tubules (short arrow).H & E, × 400

Bayero Journal of Medical Laboratory Science, BJMLS 81

Akhigbemen and Idomeh (2024) BJMLS, 9(1): 72 - 86



Figure 7: Representative photomicrographs of the heart of streptozotocin induced diabetic rats. A: positive control 5mg/kg glibenclamide, B; extract 200mg/kg, C: extract 400mg/kg, D: 800mg/kg , E:diabetic untreated, F:non-diabetic animals. Prominent myocardial fibers cells (short arrow), interstitial space and mildly enlarged coronary artery (long arrow). H & E, × 400



Figure 8: Representative photomicrographs of the spleen of streptozotocin induced diabetic rats. A: positive control 5mg/kg glibenclamide, B; extract 200mg/kg, C: extract 400mg/kg, D: 800mg/kg, E: diabetic untreated, F: non-diabetic animals. Photomicrographs shows visible dispersed lymphoid follicles (white pulp) and red pulps consisting of aggregates of lymphocytes with eccentrically located blood vessels (long arrow).H & E, $\times 400$

Bayero Journal of Medical Laboratory Science, BJMLS 82

Biochemical and Heamatological Evaluation



Figure 9: Representative photomicrographs of the pancreases of streptozotocin induced diabetic rats. A: positive control 5mg/kg glibenclamide, B; extract 200mg/kg, C: extract 400mg/kg, D: 800mg/kg, E: diabetic untreated, F:non-diabetic animals. Pancreas reveals acinar pattern structure with pyknotic nuclei of some acinar cells appearing (short arrow). The acinar cells which stained strongly are arranged in lobules with mildly congested pancreatic duct (long arrow). H & E, \times 400



Figure 10: Representative photomicrographs of the Brain of streptozotocin induced diabetic rats. A: positive control 5mg/kg glibenclamide, B; extract 200mg/kg, C: extract 400mg/kg, D: 800mg/kg, E: diabetic untreated, F: non-diabetic animals. Brain reveals molecular layer (short arrow) and granule layer (long arrow) with white matter and Purkinje cell. H & E, \times 400

DISCUSSION

This study reveals the presence of 35 compounds in the stem bark extract of bombax buonopozense which might be responsible for biologic the activity observed when the plant is employed ethno medicinally in the management of diabetes mellitus. Diabetes mellitus (DM) is a characterized metabolic disease by hyperglycemia, which if untreated can lead to long-term damage and dysfunction of vital organs like the eyes, kidneys, nerves, heart and blood vessels. Biochemical and hematological parameters are important tools for assessing the physiological profile of vital organs (Pessini et al., 2020) and by extension; are indicative of toxicity from any substances consumed.

Erythrocytes, also called red blood cells (RBCs), are the most glucose-consuming cells in the human body. In the presence of hyperglycemia, long-lasting the morphology, metabolism, and function of erythrocytes are inevitably subject to a series of changes that further affect hemorheology and microcirculation (Sprague et al., 2006; Zhou et al 2018). In the present study, the extract seems not to have had any adverse effect on the hematological parameters. White blood cell count is a marker of inflammation. Increased neutrophil and lymphocyte counts can predict Type 2 diabetes, however; this is not the case for monocyte count (Gkrania-Klotsas et al., 2010). Similarly, the concentration of white blood cell is directly associated with insulin resistance (Vozarova et al., 2002; Hanley et al., 2009) while being inversely related with insulin secretion, which is often used to predict worsening of insulin sensitivity (Hanley et al., 2009).

Electrolytes plays essential role in quite a lot of bodily functions; such as maintenance of acid-base balance, membrane potential, muscle contraction, nerve conduction and regulation of body fluid (Khan *et al.*, 2019). Any significant alterations in electrolytes homeostasis may lead to physiologic disorders. Diabetes mellitus associated hyperglycemia causes glucose induced osmotic diuresis with a resultant loss of body fluids and electrolytes (Ojiako and Chikezie, 2015). Toxicity in the kidneys can manifests as oliguria and or alteration in the plasma levels of electrolytes, urea and creatinine is one of the most significant complications of Diabetes mellitus.

Lipid profile indices are useful in monitoring the functionality or derangement in the cardiovascular system (Flegal et al., 2002). Elevated level of triglycerides and LDL-C is a predisposing factor to atherosclerosis and several cardiovascular diseases (Rishi et al., 2016; Nicholls et al., 2018; George et al., 2021). The present study has shown that serum lipids are not significantly altered. Patients with type 2 diabetes mellitus and prediabetes often experience abnormal lipid levels termed "diabetic dyslipidemia", which is characteristically described by high levels of total cholesterol triglycerides and low density lipoprotein as well as low levels of high density lipoprotein cholesterol (Mooradian 2009); Santos-Gallego and Rosenson, 2014). It is also known that low density lipoprotein cholesterol levels may be moderately increased or normal.

The liver plays essential role that include. detoxification various primary of proteins, metabolites. synthesizing and producing digestive enzymes. Several liver function tests can help determine the site of liver damaged. These tests do not denote the functional state of the liver rather they point and show the source of damage to the liver (Lala, 2023). For, instance elevation in ALT and AST compared to ALP, and bilirubin signifies a hepatocellular disease; whereas, an elevation in ALP and bilirubin when compared to ALT and AST would portray a cholestasis pattern. A mixed injury pattern on the other hand, is suggestive of an elevation in alkaline phosphatase and AST/ALT levels. Elevation of bilirubin with normal alkaline phosphatase and AST/ALT levels is suggestive of isolated hyperbilirubinemia (Vagvala and O'Connor, 2018).

CONCLUSION

Findings, from the biochemical and hematological test correlate with results from the histology of selected vital organs showing that the plant is relatively safe. In addition, the bioactive compounds as present in this compound might be responsible for its use in ethnomedicine.

REFERENCES

- Agarwal, R., Hazarika, B., Gupta, D., Agarwal, A.N. and Chakrabarti, A. (2010). Aspergillus hypersensitivity in patients with chronic obstructive pulmonary disease: COPD as a risk factor for ABPA. Medical Mycology, 48: 988-994
- Afrisham, R., Aberomand, M., Ghaffari, M.A.,, Siahpoosh, A. and Jamalan, M.(2015). Inhibitory Effect of Heracleum persicum and Ziziphus jujuba on Activity of Alpha-Amylase. *Journal of Botany*, 1–8.
- American Diabetes Association (2013). Diagnosis and classification of diabetes mellitus. *Diabetes Care*, **37**(Supplement_1):S81–S90.
- Beentje, H., and Smith, S., (2001). Plant systematic and Phytogeography for the understanding of African Biodiversity. *Systematic and Geography of plants*, **71**(1):234–286.
- Chisom, F.I., Okereke, C. and Okeke, C.U., (2014). Comparative phytochemical and proximate Analyses on Ceiba pentandra (L) Gaertn and Bombax buonopozense (P) Beauv. *International Journal of Herbal Medicine*, 2(2):162-167
- Danso, J., Alemawor, F., Boateng, R., Barimah, J. and Kumah, D. B. (2019). Effect of drying on the nutrient and anti- nutrient composition of Bombax buonopozense sepals. *African Journal of Food Science*, **13**(1), 21-29.
- Gkrania-Klotsas, E., Ye. Z. and Cooper, A.J. (2010). Differential white blood cell count and type 2 diabetes: systematic

Conflict of Interest

The authors declare no conflict of interest.

Acknowledgments

The authors are grateful to Mr. Ibeh, Vote Samson and Seyi Omoluyi and the entire Laboratory staff of the Department of Pharmacology & Toxicology, University of Benin for their assistance.

> review and meta-analysis of crosssectional and prospective studies. *Public library of science One*, **5**:e13405

- Hanley, A.J, Retnakaran, R. and Qi, Y. (2009). Association of hematological parameters with insulin resistance and beta-cell dysfunction in nondiabetic subjects. *Journal of Clinical Endocrinology Metabolism*, 94:3824–3832.
- Kooti, W., Farokhipour, M., Asadzadeh, Z., Ashtary-Larky, D. and Asadi-Samani, M. (2016). The role of medicinal plants in the treatment of diabetes: a systematic review. Electron Physician, 15; 8(1):1832-42.
- Khan, R. N., Saba, F., Kausar, S. F., and Siddiqui, M. H. (2019). Pattern of electrolyte imbalance in Type 2 diabetes patients: Experience from a tertiary care hospital. *Pakistan Journal of Medical Sciences*, 35(3), 797-801.

V., Zubair, M. and Minter, D.A. Lala, (2023)Liver Function Tests. [Updated 2023 Jul 30]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing. Mooradian, A.D (2009).Dyslipidemia in type 2 diabetes mellitus. Nature Clinical Practice Endocrinology and Metabolism, **5**:150–159.

Ojiako, O.A. and Chikezie, P.C. (2015). Blood Na±/K±and Cl levels of Hyperglycemic rats administered with traditional herbal formulations. *Pharmacognosy Communications*, **5**(2):140–144.

- Pessini, P.G.D.S., Knox de Souza, P.R. Chagas, C.D.S, Sampaio, E.G., Neves, D.S., Petri, G., Fonseca, F.L.A. and da Silva, E.B. (2020). Hematological reference values and animal welfare parameters of BALB/C-FMABC (*Mus musculus*) inoculated with Ehrlich tumor kept in ABC Medical the vivarium at Model School. Animal and Experimental Medicine, 29:3(1):32-39.
- Santos-Gallego, C.G. and Rosenson, R.S.(2014). Role of HDL in those with diabetes. *Current Cardiology Reports*, 16:512
- Sprague, R. S., Stephenson, A. H., Bowles,E. A., Stumpf, M. S. and Lonigro, A.J.(2006). Reduced expression of Giin erythrocytes of humans with type2 diabetes is associated with

impairment of both cAMP generation and ATP release. *Diabetes*. **55**(12):3588–3593.

- Vagvala, S.H. and O'Connor, S.D.(2018). Imaging of abnormal liver function tests. Clin Liver Dis (Hoboken). **11**(5):128-134.
- Vozarova, B., Weyer, C., Lindsay, R.S., Pratley, R.E, Bogardus, C. and Tataranni, P.A.,(2002). High white blood cell count is associated with a worsening of insulin sensitivity and predicts the development of type 2 diabetes. *Diabetes*, **51:455**–461.
- Zhou, Z., Mahdi, A.and Tratsiakovich, Y. (2018). Erythrocytes From Patients With Type 2 Diabetes Induce Endothelial Dysfunction Via Arginase I. Journal of the American College of Cardiology.**72** (7):769– 780.