International Journal of Global Affairs, Research and Development (IJGARD), Vol.4, No.1, 2026, 599-628, ISSN 2992-2488

ANTIDIABETIC, ANTIOXIDANT, AND PROTECTIVE EFFECTS OF AQUEOUS EXTRACTS OF *SORGHUM BICOLOR* AND *PENNISETUM GLAUCUM* SEEDS IN ALLOXAN-INDUCED DIABETIC WISTAR RATS

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ABSTRACT

Diabetes mellitus is a chronic metabolic disorder characterized by persistent hyperglycemia and associated metabolic complications resulting from impaired insulin secretion, insulin resistance, or both. Despite the increasing global burden of diabetes, especially in low- and middle-income countries, many medicinal plants and cereal grains used traditionally for diabetes management remain scientifically underexplored. Sorghum bicolor and Pennisetum glaucum are widely consumed cereal crops rich in bioactive phytochemicals with potential therapeutic properties. The absence of scientific data, to the best of our knowledge, on the age long folkloric use of sorghum and millet at 750 and 1000 mg/kg body weight by sufferers of diabetes prompted the present investigation This study investigated the antidiabetic, antioxidant, and protective effects of aqueous extracts of Sorghum bicolor and Pennisetum glaucum seeds in alloxan-induced diabetic Wistar rats.Forty nine albino rats of both sexes were divided into seven groups (A-G) of 7 rats each. Animals in group A (control) received distilled water while those in groups B-G which were induced into hyperglycemia by intraperitoneal injection of alloxan monohydrate (150 mg/kg body weight) received distilled water, 25 mg/kg body weight of glibenclamide (reference antidiabetic drug), 750 mg/kg body weight of S. bicolor, 1000 mg/kg body weight of S. bicolor, 750 mg/kg body weight of P. glaucum and 1000 mg/kg body weight of P. glaucum respectively. Extract administration of 0.5 ml for was done once daily for 21 days after which blood glucose level, selected biochemical parameters and pancreatic histology were determined. Alloxan induction significantly ($p<0.05$) increased the levels of blood glucose (BG), malonaldehyde (MDA), total cholesterol (TC), triglyceride (Trig), low-density lipoprotein cholesterol (LDLC), urea, uric acid, creatinine as well as activities of alkaline phosphatase (ALP), aspartate aminotransferase (AST), alanine amino transferase (ALT) in the serum of the animals. Administration of alloxan significantly ($p<0.05$) decreased the levels of high-density lipoprotein cholesterol (HDLC) as well as the activities of catalase (CAT) and superoxide dismutase (SOD) in the serum of the animals. However, when compared with the distilled water treated diabetic animals, treatment of hyperglycaemic/diabetic animals with the aqueous extract of both S. bicolor and P. glaucum seed at 750 and 1000 mg/kg body weight which significantly ($p<0.05$) decreased the levels of BG, MDA, TC, Trig, LDLC, urea, uric acid, creatinine as well as activities of ALP, AST, ALT, HDLC as well as activities of CAT and SOD in the serum of the animals, in a manner comparable with the glibenclamide treated diabetic animals. Histological damage in the pancreas following alloxan administration was restored in diabetic animals treated with both aqueous extract of S. bicolor and P. glaucum seed at 750 and 1000 mg/kg body weight in a manner comparable with the glibenclamide treated diabetic animals The findings of this study demonstrate that aqueous

extracts of Sorghum bicolor and Pennisetum glaucum possess significant antidiabetic, antioxidant, and protective activities against alloxan-induced hyperglycemia and associated metabolic dysfunctions. The study scientifically supports the traditional use of these cereal grains in the management of diabetes mellitus, with Pennisetum glaucum at 1000 mg/kg body weight showing the greatest therapeutic potential.

Keywords: Diabetes mellitus; *Sorghum bicolor; Pennisetum glaucum;* oxidative stress; antioxidant activity; histopathology; alloxan; hyperglycemia.

1.0 INTRODUCTION

Diabetes mellitus is one of the most prevalent non-communicable diseases (NCDs) worldwide and remains a major public health challenge due to its increasing prevalence and associated complications. The disease is characterized by chronic hyperglycemia resulting from impaired insulin secretion, insulin resistance, or both, leading to disturbances in carbohydrate, lipid, and protein metabolism (World Health Organization [WHO], 2023). According to the International Diabetes Federation (IDF), approximately 537 million adults were living with diabetes globally in 2021, with projections indicating a rise to 783 million by 2045 (International Diabetes Federation [IDF], 2021). Recent reports further suggest that the global burden of diabetes will continue to increase substantially, particularly in developing regions such as Africa, Asia, and the Middle East (Genitsaridi *et al*., 2026; Sun *et al*., 2022).

Persistent hyperglycemia contributes to the development of several microvascular and macrovascular complications, including diabetic nephropathy, neuropathy, retinopathy, coronary artery disease, stroke, and cardiovascular disorders (Low Wang *et al*., 2016). In addition, diabetes is commonly associated with dyslipidemia, hypertension, hypercholesterolemia, and oxidative stress, all of which contribute significantly to disease progression and tissue damage (Geijselaers *et al*., 2017). Oxidative stress plays a critical role in the pathogenesis of diabetes through excessive production of reactive oxygen species (ROS), leading to lipid peroxidation, inflammation, mitochondrial dysfunction, and cellular injury (Asmat *et al*., 2016).

The increasing burden of diabetes has intensified the search for safer and affordable therapeutic agents from natural sources. Medicinal plants and functional foods rich in phytochemicals have attracted considerable scientific interest because of their antioxidant and antihyperglycemic properties (Aderibigbe *et al*., 2021). Among these, Sorghum bicolor and Pearl millet (*Pennisetum glaucum*) are important cereal crops widely cultivated in Africa and Asia and are recognized for their nutritional and therapeutic potential.

Sorghum bicolor contains bioactive compounds such as phenolic acids, flavonoids, tannins, and anthocyanins, which possess antioxidant and antidiabetic activities (Awika & Rooney, 2004). Previous studies have shown that sorghum-derived polyphenols may improve glucose metabolism through inhibition of α-amylase and α-glucosidase activities, reduction of glucose absorption, and enhancement of insulin sensitivity (Stefoska-Needham *et al*., 2015). Similarly, *Pennisetum glaucum* is rich in dietary fiber, minerals, polyphenols, and phytates, which have been reported to exhibit antioxidant, hypolipidemic, and antihyperglycemic effects (Anitha *et al*., 2017; Sharma *et al*., 2021). Dietary polyphenols present in millet may regulate postprandial glycemic response and reduce oxidative stress associated with diabetes mellitus (Bathaie & Mousavi, 2018).

Despite the documented nutritional and pharmacological benefits of sorghum and pearl millet, limited information exists regarding the protective effects of their aqueous seed extracts against diabetes-induced metabolic abnormalities. Therefore, the present study investigated the antidiabetic, antioxidant, and protective effects of aqueous extracts of *Sorghum bicolor* and *Pennisetum glaucum* seeds in alloxan-induced diabetic Wistar rats.

2.0 MATERIALS AND METHODS

2.1 Plant Materials and Authentication

Plant materials were collected by the researcher at Karu, Nasarawa State, Nigeria. The samples were subsequently transported to the National Cereals Research Institute (NCRI), Badeggi, Bida, Niger State, Nigeria, for authentication.

2.2 Experimental Animals

Adult Wistar rats (*Rattus norvegicus*) were purchased from the National Veterinary Research Institute (NVRI), Vom, Plateau State, Nigeria. The animals were maintained under standard laboratory conditions prior to experimentation. They were housed in well-ventilated cages, fed with standard pellet diet, and provided with water *ad libitum*. The rats were allowed a 14-day acclimatization period before the commencement of the study.

2.3 Assay Kits, Chemicals, and Drugs

Assay kits for the determination of liver function, kidney function, lipid profile, selected biomolecules, and carbohydrate-metabolizing enzymes were obtained from Randox Laboratories Ltd., Co. Antrim, UK.

Alloxan monohydrate (1,3-diazinane-2,4,5,6-tetrone) and glibenclamide were purchased from Kemei Laboratories Ltd., Mumbai, India, and May & Baker Ltd., Dagenham, England, respectively.

Other chemicals used included normal saline, glacial acetic acid, o-toluidine reagent, sulfuric acid, catalyst mixture, distilled water, sodium hydroxide, boric acid, hydrochloric acid, petroleum ether, trichloroacetic acid, nitric acid, absolute ethanol, chloroform/ethanol mixture, acetate buffer, ammonium thiocyanate, ferric chloride solution, potassium permanganate, oxalic acid, methyl red indicator, and calcium chloride.

2.4 Glucometer and Test Strips

Fasting blood glucose levels were measured using the OneTouch Blood Glucose Monitoring System (Schiffgraben Diagnostic, Hannover, Germany) with On-Call Redi blood glucose test strips (ACON Laboratories Inc., San Diego, USA).

2.5 Preparation of Aqueous Extract of *Sorghum bicolor* and *Pennisetum glaucum*

Seeds were first ground into coarse form using a mortar and pestle and further milled into fine powder. Each powdered plant material (150 g) was separately soaked in 500 mL of distilled water for 6 hours with constant stirring. The resulting mixtures were centrifuged, and the supernatants were collected. The extracts were concentrated using a water bath and subsequently dried at room temperature. The dried extracts were weighed, stored in sterile specimen containers, and used for preparation of stock solutions for administration.

2.6 Induction of Hyperglycemia and Antidiabetic Study Design

Hyperglycemia was induced in Wistar rats via a single intraperitoneal injection of freshly prepared alloxan monohydrate solution at a dose of 150 mg/kg body weight, following an overnight fast of 12 hours.

2.7 Confirmation of Hyperglycemia

The induction of hyperglycemia was confirmed 72 hours post-alloxan administration by measuring fasting blood glucose levels. Rats with fasting blood glucose levels between 360–590 mg/dL were considered diabetic and included in the study.

2.8 Animal Grouping and Extract Administration

The animals were randomly divided into seven groups as follows:

- Group A: Normal control (received feed and water only)
- Group B: Diabetic control (received distilled water)
- Group C: Diabetic + glibenclamide
- Group D: Diabetic + 750 mg/kg body weight *Sorghum bicolor* extract
- Group E: Diabetic + 1000 mg/kg body weight *Sorghum bicolor* extract
- Group F: Diabetic + 750 mg/kg body weight *Pennisetum glaucum* extract
- Group G: Diabetic + 1000 mg/kg body weight *Pennisetum glaucum* extract

2.9 Blood Collection, Serum Preparation, and Tissue Processing

After 21 days of treatment, body weights were recorded. Blood samples were collected via retro-orbital venous plexus puncture into plain sample tubes for biochemical analysis. The blood samples were centrifuged to obtain serum, which was stored in labeled plain bottles for further assays.

Harvested organs were excised and preserved in formol saline for histopathological analysis.

2.10 Determination of serum enzymic and non-enzymic antioxidant activity Determination of Serum insulin

Determination of malonaldehyde (MDA) concentration

Malondialdehyde (MDA), an index of lipid peroxidation was determined in the serum using the method of Buege and Aust (1978). 1.0 ml of the appropriately diluted sample was added to 2 ml of MDA reagent and boiled at 100 ^{0}C for 15 minutes. The reaction mixture was then allowed to cool down. The flocculent materials were removed by centrifugation at 3000 g for 10 minutes. The supernatant was removed and its absorbance was read at 532 nm against a blank. The concentration of MDA was calculated from the expression:

Concentration of MDA= $\Delta A \times TV / \varepsilon \times SV$

Where:

ΔA= change in absorbance

TV= total volume

SV= sample volume

ε = molar extinction = $1.56 \times 10^5\ M^{-1}cm^{-1}$

Determination of Seperoxide dismutase (SOD)

The superoxide dismutase activity in pancreas and liver supernatant was determined by the method described by Misra and Fridovich (1972). To 0.2 ml of appropriately diluted tissue supernatant, 2.5 ml of 0.05 M carbonate buffer (pH 10.2) was added. The reaction was initiated by adding 0.3 ml of freshly prepared 0.3 mM of epinephrine (substrate) and 0.2 ml of distilled water. This was mixed by inversion. The reference cuvette contained 2.5 ml of the buffer, 0.2 ml of distilled water and 0.3 ml of epinephrine. The increase in absorbance was read at 480 nm at interval of 30 seconds for 150 seconds.

Calculation

% Inhibition = Increase in absorbance of substrate

Determination of Catalase (CAT) activity

Catalase was assayed in the serum by the procedure of Beers and Sizer (1952). 2.9 ml of 0.036% w/w hydrogen peroxide was added and mixed with 0.1 ml of appropriately diluted tissue homogenate. A blank was prepared containing potassium phosphate buffer (50 nM, pH 7.0, 3.0 ml). The time required for the $A_{240\ nm}$ of the reaction mixture in step 1 and 2 to decrease from 0.45

to 0.40 absorbance units was recorded. The initial $A_{240\ nm}$ exceeded 0.450 absorbance units. The activity and specific activity of catalase was calculated using the expression:

$$\text{Enzyme Activity (nmol/min/ml)} = \frac{3.45 \times 1000 \times DF}{Min \times V}$$

Where:

3.45= correspond to the decomposition of 3.45 micromoles of hydrogen peroxide in a 3.0 ml reaction mixture producing a decrease in the $A_{240\ nm}$ from 0.45 to 0.40 absorbance units.

DF= Dilution factor

Determination of serum liver function indices

Determination of alkaline phosphatase (ALP) activity

The procedure described by Wright *et al* (1972b) was employed for the determination of alkaline phosphatase activity. Two test tubes were labelled blank and test. 2.2ml of 0.1M of carbonate buffer was added poured tubes, then 0.1 M of $MgSO_4.7H_2O$ was added and then 0.2ml of distilled water was added to the blank while 0.2ml of enzyme source was added to the test. Both tubes were incubated at 37^0C for 10 minutes. 0.5ml of P-nitrophenyl phosphate (10mM) was then added to both tubes and incubated at 37^0C for 30minutes. The reaction was terminated with 2.0ml of 1M sodium hydroxide.

The absorbance was read spectrophotometrically at 400 nm. Enzyme activity was calculated using the following expression:

$$\text{Enzyme activity (nM/min/ml)} = \frac{AB/min \times 1000 \times TV \times F}{9.9 \times SV \times L}$$

Where:

AB/min = Absorbance of reaction mixture per minute

TV = Total volume of the reaction mixture.

F = Total dilution factor

SV = Volume of enzyme source

L = Light path length (1cm)

9.9 = Extinction co-efficient of 1 μm of p-nitrophenol in an alkaline solution of 1 ml and 1 cm path length

1000 = The factor introduced to enable the enzyme activity to be expressed in nM/min/mg protein.

The specific activity for alkaline phosphatase was calculated from the expression:

$$\text{Specific enzyme activity (nM/min/mg protein)} = \frac{\text{Enzyme activity}}{\text{Protein concentration}}$$

Determination of aspartate aminotransferase (AST) activity

The procedure described by Reitman and Frankel (1957) was used for assaying the activity of aspartate aminotransferase. Two test tubes were labelled blank and test. 0.1ml of the sample was poured in the test tube labelled test. 0.5ml of reagent 1 was added to both test tubes. 0.1ml of distilled water was then added to the blank. It was mixed and incubated for 30 minutes at 37^0C. 0.5ml of reagent 2 was added to both test tubes. It was then mixed and allowed o stand for 20minutes at 25^0C then 0.5ml of sodium hydroxide was then added to both test tubes. The solution was mixed and absorbance read against blank after 5 minutes at 468 nm. The enzyme activity was obtained from the calibration curve.

Determination of alanine aminotransferase (ALT)

The procedure described by Reitman and Frankel (1957) was used for assaying the activity of alanine aminotransferase. Two test tubes were labelled blank and test. 0.1ml of the sample was poured in the test tube labelled test. 0.5ml of reagent 1 was added to both test tubes. 0.1ml of distilled water was then added to the blank. It was mixed and incubated for 30 minutes at 37^0C. 0.5ml of reagent 2 was added to both test tubes. It was then mixed and allowed to stand for 20minutes at 25^0C then 0.5ml of sodium hydroxide was then added to both test tubes. The solution was mixed and absorbance read against blank after 5 minutes at 468 nm. The enzyme activity was obtained from the calibration curve.

Determination of serum kidney function parameters

Determination of Urea concentration

The diacetyl monoxime method described by Rosenthal (1955) was used for the determination of urea in serum of the animals. Test tubes were set and labeled B (Blank), STD (Standard) and TS (Test Sample) accordingly. A known amount (9.9 ml) of distilled water was pipetted into STD and TS while 0.1 ml of the serum sample was pipetted into STD and TS. This was mixed thoroughly, and 1.0 ml of it was pipetted into TS. 2.0 ml of distilled water was pipetted into B. 2 ml of mixed coloured reagent was added to each of the test tubes while 2 ml of mixed acid reagent was also added to each of the test tubes. The solution was mixed thoroughly in water bath at 40 ^{0}C for 20 minutes, allowed to cool and read spectrophotometrically at 520 nm.

Calculation

$$\text{Urea concentration (mg/100 ml)} = \frac{\text{Absorbance of Sample x Concentration of Standard x 100}}{\text{Absorbance of Standard x Volume of Sample}}$$

Determination of Serum Creatinine

The method described by Jaffe (1941) was adopted for the determination of creatinine in the serum of the animals. The determination of creatinine was divided into two stages. Stage I involves addition of 2 ml of serum and 2ml of distilled water. This was mixed, allowed to stand for 5 minutes followed by addition of 3 ml of 5% sodium tungstate and 2 ml of $_{2/3}NH_2SO_4$. This was centrifuge at 2000 g for 10 minutes. At stage II, 3 ml of supernatant from stage I was added to each of the test tubes. Test tubes were set and labeled TS (Test Sample), STD (Standard) and B (Blank) accordingly. Three ml of serum, creatinine standard (1 mg/100 ml) and distilled water were pipetted into TS, STD and B respectively. 1 ml of 0.04M picric acid was added to each of the test tubes, while 1 ml of 0.75 NaOH was also added to each of the test tubes. The solution was mixed after each addition and left at 25 ^{0}C for 15 minutes. Absorbance was read spectrophotometrically at 540 nm.

Calculation

$$\text{Creatinine Conc. (mg/100 ml)} = \frac{\text{Absorbance of Sample x Concentration of Standard x 100}}{\text{Absorbance of Standard x Volume of Sample}}$$

Determination of uric acid concentration

Serum uric acid was determined according to the procedure described by Tietz (1995). Three test tubes were labeled test, blank and standard. 0.5ml of distilled water was added to the standerd test tube while 0.5ml of sample was poured in the test tube labeled test. 0.5ml of sulphuric acid was added to both test tubes and 0.5ml of sodium tunsgate was then added. The solutuin was mixed and centrifuged. 3ml of the standard was poured in the test tube labeled blank. 0.5ml of 14% Na_2CO_3 was added to all the test tubes and then 0.5ml of tungistic acid was finally added. The solution was left undisturbed for 20 minutes at room temperature for the blue colour to develop after which the absorbance was read at wavelength of 620 nm.

Calculation

$$\text{Concentration of uric acid (mg/dl)} = \frac{A_{sample}}{A_{standard}} \times \text{Concentration of standard}$$

Concentration of standard = 10.48 mg/dl

Determination of serum lipids

Determination of total cholesterol concentration

The determination of the concentration of total cholesterol in the serum of the animal was done using the "CHOD-PAP" reaction described by Fredrickson *et al* (1967). Test tubes were set and labeled B (Blank), TS (Test Sample) and STD (Standard) accordingly. A known amount (0.02 ml) of distilled water, serum sample and standard were pipetted into B, TS and STD test tubes respectively. 2 ml of working reagent was added to each of the test tubes (Blank, Test Sample and Standard), mixed and incubated at 25 ^{0}C for 5 minutes. The solutions were mixed properly, incubated at 25 ^{0}C for 10 minutes and the absorbance of the test sample (A_{sample}) and standard ($A_{standard}$) was read the reagent blank within 60 minutes at 546 nm.

Calculation

$$\text{Concentration of Cholesterol (mmol/L)} = \frac{\text{Absorbance of Sample x Concentration of Standard}}{\text{Absorbance of Standard}}$$

Concentration of Standard = 5.10 mmol/L

Determination of triglyceride concentration

Triacylglycerol was determined in serum of the animals using the glycerol phosphate oxidase reaction described by Tietz (1990). This involved setting test tubes according to B (Blank), STD (Standard) and TS (Test sample). Ten micro litres each of distilled water, standard and test sample were pipetted into test labeled B, STD and TS respectively. Later, 1000 µl of the working reagent was pipetted into all the test tubes: B, STD and TS. The test tube contents were mixed thoroughly and incubated for 10 minutes at 25 ^{0}C. The absorbance of the Sample (A_{sample}) and Standard ($A_{standard}$) were read against the reagent blank at 546 nm wavelength. Finally, the concentration of triacylglycerol in the serum was determined.

Calculation

$$\text{Triacylglycerol Concentration (mmol/L)} = \frac{\text{Absorbance of Sample x Concentration of Standard}}{\text{Absorbance of Standard}}$$

Concentration of Standard = 2.19 mmol/L

Determination of low density lipoprotein cholesterol (LDL) concentration

Serum LDL-cholesterol was calculated using the Friedewald formula (Friedewald *et al*., 1972). LDL cholesterol concentration in the sample was calculated using the expression:

$$\text{LDL cholesterol conc. (mmol/L)} = \text{Total cholesterol} - \frac{\text{Triacylglycerol}}{} - \text{HDL cholesterol}$$

2.2

Determination of high density lipoprotein cholesterol (HDL) concentration

HDL-cholesterol was determined in the serum of the animals by adopting the procedure described by Hainline *et al* (1980). 500 µl of precipitating solution (phosphotungstic acid in the presence of magnesium ions) was added to 200 µl of the sample while addition of 200 µl constituted standard reagent. These were properly mixed and allowed to stand for 15 minutes at room temperature, followed by centrifugation of the resulting supernatant at 4000 g for 10 minutes. The HDL-cholesterol present in the supernatant was finally determined spectrophotometrically at 546 nm.

Calculation

$$\text{Concentration of HDL chol. (mmol/L)} = \frac{\text{Absorbance of Sample x Concentration of Standard}}{\text{Absorbance of Standard}}$$

Concentration of Standard = 5.10 mmol/L

2.11 Data analysis

The data was analyzed using SPSS. The data was expressed as Mean ± Standard Error of Mean (SEM) of seven determinations. The group means was compared by Newman-Keuls multiple range test (NKMRT). Values was considered statistically significant at $p<0.05$.

3.0 RESULTS AND DISCUSSION

3.1 Results

3.11 Blood glucose level of alloxan-induced hyperglycemic rats following oral administration of aqueous *Sorghum bicolor* and *Pennisetum glaucum* seed

The results showed that all the animals treated with 150 mg/kg body weight of alloxan became diabetic after 72 hours with blood glucose level ranging from 302.96±0.83 to 491.42±2.58 mg/dL (Table 1). The fasting blood glucose level of the animals administered with alloxan and distilled water significantly ($p< 0.05$) increased from 92.38±0.75 to 599.34±3.87 mg/dL. Administration of the aqueous extract of *Sorghum bicolor* and *Pennisetum glaucum* seed both at the doses of 750 and 1000 mg/kg body weight significantly ($p< 0.05$) reduced the blood glucose levels progressively up till the end of the treatment period. While the reduction in fasting blood glucose of alloxan-induced diabetic animals administered 1000 mg/kg body weight of *Pennisetum glaucum* aqueous extract produced values that compared ($p> 0.05$) well with the distilled water treated non-diabetic animals on day 21, the fasting blood glucose level of alloxan-induced diabetic animals administered 750 and 1000 mg/kg body weight of both *Sorghum bicolor* and *Pennisetum glaucum*

aqueous extract, although, reduced, did not compare well with the distilled water treated non-diabetic animals from Day 1 to Day 21 (Table 1).

Table 1: Blood glucose level (mg/dL) of alloxan-induced diabetic rats following oral administration of aqueous extract (750 and 1000mg/kg body weight) of *Sorghum bicolor* and *Pennisetum glaucum*

			Treatment Days			
Treatment Group	Basal BG	BG Prior to Treat.	Day 4	Day 8	Day 12	Day 14 Day 18 Day 21
A	83.42±0.42[a]	75.27±0.12[a]	82.48±0.02[a]	78.56±0.23[a]	70.68±0.75[a]	72.36±0.46[a] 86.24±0.34[a] 80.05±0.89[a]
B	92.38±0.75[b]	413.67±1.88[b]	441.02±1.58[b]	507.45±2.58[b]	513.04±2.16[b]	529.47±2.78[b] 547.23±3.05[b] 599.34±3.87[b]
C	82.84±0.37[a]	491.42±2.58[c]	416.13±1.34[c]	325.46±1.87[c]	274.17±1.56[c]	163.58±1.89[c] 103.67±0.56[c] 72.13±0.96[c]
D	81.35±0.03[a]	366.67±0.83[d]	316.53±1.75[d]	278.49±1.34[d]	223.14±1.72[d]	155.67±1.25[d] 121.53±0.36[d] 96.03±0.86[d]
E	85.46±0.56[c]	425.67±1.46[e]	339.04±1.58[e]	281.53±1.68[e]	202.45±1.73[e]	171.68±2.06[e] 124.67±1.52[e] 87.16±0.74[e]
F	72.94±0.79[d]	302.96±0.83[f]	261.48±0.28[f]	215.76±1.74[f]	172.38±1.36[f]	154.89±2.73[f] 129.73±0.14[f] 99.05±0.35[d]
G	76.39±0.84[d]	405.83±1.38[g]	314.17±1.75[g]	281.63±1.62[e]	188.45±1.35[g]	145.32±2.86[g] 115.38±0.67[g] 81.32±0.78[a]

A = Control; **B** = Diabetic rats + Distilled water; **C =** Diabetic rats + Glibenclamide; **D** = Diabetic rats + 750mg/kg body weight of *Sorghum bicolor* Extract; **E =** Diabetic rats + 1000 mg/kg body weight of *Sorghum bicolor* Extract; **F =** Diabetic rats + 750 mg/kg body weight of *Pennisetum glaucum* Extract; **G** = Diabetic rats + 1000 mg/kg body weight of *Pennisetum glaucum* Extract.

Data are mean ± SEM of seven determinations. Test values with superscript different from their respective control down the column for each day are significantly different ($p<0.05$)

3.1.2 Serum antioxidant status of alloxan-induced hyperglycemic rats following oral administration of aqueous extract of *Sorghum bicolor* and *Pennisetum glaucum* seed

Administration of alloxan significantly (P<0.05) increased serum malonaldehyde concentration when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) decreased serum malonaldehyde concentration when compared with distilled water treated hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor* and *Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) decreased serum malonaldehyde concentration when compared with distilled water treated hyperglycemic animals (Table 2).

Administration of alloxan significantly (P<0.05) decreased serum superoxide dismutase activity when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) increased serum superoxide dismutase activity when compared with distilled water treated hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) increased serum superoxide dismutase activity when compared with distilled water treated hyperglycemic animals (Table 2).

Administration of alloxan significantly (P<0.05) decreased serum catalase (CAT) activity when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) increased serum catalase (CAT) activity when compared with distilled water treated diabetic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) increased serum catalase (CAT) activity when compared with distilled water treated hyperglycemic animals (Table 2).

Table 2: Serum Antioxidant Status of Alloxan-induced Diabetic Rats Following Oral Administration of Aqueous Extract of *Sorghum bicolor* and *Pennisetum glaucum* Seed

Treatment Group	Catalase (CAT) (U/mg Protein)	Superoxide Dismutase (SOD) (U/mg Protein)	Malondialdehyde (MDA) (U/mg Protein)
A	1286.88±2.24[a]	238.75±1.04[a]	4.35±0.07[a]
B	208.71±0.98[b]	30.98±0.08[b]	10.51±0.52[b]
C	507.11±1.72[c]	64.97±0.42[c]	4.68±0.09[a, c]
D	346.40±1.36[d]	82.95±0.50[d]	6.53±0.11[d]
E	4571.38±1.09[e]	104.13±0.75[e]	5.66±0.10[e]
F	724.91±1.76[e]	143.77±0.87[f]	6.38±0.12[d, f]
G	902.63±1.87[f]	188.81±0.98[g]	8.37±0.14[g]

Data are mean ± SEM of seven determinations. Values with superscript different from their respective control down the column are significantly different ($p<0.05$)
A = Control; **B** = Diabetic rats + Distilled water; **C =** Diabetic rats + Glibenclamide; **D** = Diabetic rats + 750mg/kg body weight of *Sorghum bicolor* Extract; **E =** Diabetic rats + 1000 mg/kg body weight of *Sorghum bicolor* Extract; **F =** Diabetic rats + 750 mg/kg body weight of *Pennisetum glaucum* Extract; **G** = Diabetic rats + 1000 mg/kg body weight of *Pennisetum glaucum* Extract.

3.1.3 Liver function indices of alloxan-induced hyperglycemic rats following oral administration of aqueous extract (750 and 1000mg/kg body weight) of *Sorghum bicolor* and *Pennisetum glaucum* seed

Administration of alloxan significantly (P<0.05) increased serum alkaline phosphatase (ALP) concentration when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) decreased serum alkaline phosphatase (ALP) concentration activity when compared with distilled water treated hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) decreased serum alkaline phosphatase (ALP) concentration when compared with distilled water treated hyperglycemic animals (Table 3).

Administration of alloxan significantly (P<0.05) increased serum aspartate transaminase (AST) concentration when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) decreased serum aspartate transaminase (AST) concentration activity when compared with distilled water treated

hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) decreased serum aspartate transaminase (AST) concentration when compared with distilled water treated hyperglycemic animals (Table 3).

Administration of alloxan significantly (P<0.05) increased serum alanine transaminase (ALT) concentration when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) decreased serum alanine transaminase (ALT) concentration activity when compared with distilled water treated hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) decreased serum alanine transaminase (ALT) concentration when compared with distilled water treated hyperglycemic animals (Table 3).

Table 3: Liver function indices of alloxan-induced hyperglycemic rats following oral administration of aqueous extract of *Sorghum bicolor* and *Pennisetum glaucum* seed

Treatment Group	ALP (IU/L)	AST (IU/L)	ALT (IU/L)
A	65.62±1.64[a]	42.48±2.12[a]	2.09±0.03[a]
B	120.59±2.45[c]	48.04±2.57[b]	14.59±1.28[d]
C	66.72±1.56[a]	43.82±1.26[a]	2.56±0.08[a]
D	67.46±1.81[b]	42.14±1.48[a]	5.89±0.12[b]
E	66.12±2.43[a]	43.21±1.45[a]	10.76±0.12[c]
F	68.72±1.04[b]	42.76±2.02[a]	5.06±0.06[b]
G	66.25±2.75[a]	43.35±1.85[a]	9.81±1.08[c]

Data are mean ± SEM of seven determinations. Test values with superscript different from their respective control down the column are significantly different (p<0.05)

A = Control; **B** = Diabetic rats + Distilled water; **C =** Diabetic rats + Glibenclamide; **D** = Diabetic rats + 750mg/kg body weight of *Sorghum bicolor* Extract; **E =** Diabetic rats + 1000 mg/kg body weight of *Sorghum bicolor* Extract; **F =** Diabetic rats + 750 mg/kg body weight of *Pennisetum glaucum* Extract; **G** = Diabetic rats + 1000 mg/kg body weight of *Pennisetum glaucum* Extract.

3.1.4 Kidney function indices of alloxan-induced hyperglycemic rats following oral administration of aqueous extract (750 and 1000mg/kg body weight) of *Sorghum bicolor* and *Pennisetum glaucum* seed

Administration of alloxan significantly (P<0.05) increased serum Urea concentration when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) decreased serum urea concentration activity

when compared with distilled water treated hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) decreased serum urea concentration when compared with distilled water treated hyperglycemic animals (Table 4).

Administration of alloxan significantly (P<0.05) increased serum Uric acid concentration when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) decreased serum uric acid concentration activity when compared with distilled water treated hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) decreased serum uric acid concentration when compared with distilled water treated hyperglycemic animals (Table 4).

Administration of alloxan significantly (P<0.05) increased serum creatinine concentration when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) decreased serum creatinine concentration activity when compared with distilled water treated hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) decreased serum creatinine concentration when compared with distilled water treated hyperglycemic animals (Table 4).

Table 4: Kidney function indices of alloxan-induced hyperglycemic rats following oral administration of aqueous extract of *Sorghum bicolor* and *Pennisetum glaucum* seed

Treatment Group	Urea (mg/dL)	Uric Acid (mg/dL)	Creatinine (mg/dL)
A	21.54±1.10^{a}	0.58±0.01^{a}	2.85±0.02^{a}
B	41.43±1.02^{b}	6.47±0.39^{b}	10.28±1.05^{b}
C	20.64±1.12^{a}	2.80±0.06^{c}	2.14±0.10^{a}
D	23.10±1.24^{a}	4.52±0.11^{d}	4.38±0.18^{c}
E	21.34±1.34^{a}	3.11±0.09^{d}	4.02±0.06^{c}
F	21.46±0.89^{a}	3.52±0.12^{d}	1.04±0.01^{a}
G	19.34±1.21^{a}	2.51±0.05^{c}	5.24±0.08^{c}

Data are mean ± SEM of seven determinations. Test values with superscript different from their respective control down the column are significantly different (p<0.05)

A = Control; **B** = Diabetic rats + Distilled water; **C =** Diabetic rats + Glibenclamide; **D** = Diabetic rats + 750mg/kg body weight of *Sorghum bicolor* Extract; **E =** Diabetic rats + 1000 mg/kg body weight of

Sorghum bicolor Extract; **F =** Diabetic rats + 750 mg/kg body weight of *Pennisetum glaucum* Extract; **G** = Diabetic rats + 1000 mg/kg body weight of *Pennisetum glaucum* Extract.

3.1.5 Serum lipids of alloxan-induced hyperglycemic rats following oral administration of aqueous extracts (750 and 1000mg/kg body weight) of *Sorghum bicolor* and *Pennisetum glaucum* seed

Administration of alloxan significantly (P<0.05) increased serum total cholesterol concentration when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) decreased serum total cholesterol concentration activity when compared with distilled water treated hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) decreased serum total cholesterol concentration when compared with distilled water treated hyperglycemic animals (Table 5).

Administration of alloxan significantly (P<0.05) increased serum triglycerides concentration when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) decreased serum triglycerides concentration activity when compared with distilled water treated hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) decreased serum triglycerides concentration when compared with distilled water treated hyperglycemic animals (Table 5).

Administration of alloxan significantly (P<0.05) decreased serum high density lipoprotein cholesterol (HDLC) concentration when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) increased serum high density lipoprotein cholesterol (HDLC) concentration activity when compared with distilled water treated hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) increased serum high density lipoprotein cholesterol (HDLC) concentration when compared with distilled water treated hyperglycemic animals (Table 5).

Administration of alloxan significantly (P<0.05) increased serum low density lipoprotein cholesterol (LDLC) concentration when compared with Distilled water treated control animals. Administration of glibenclamide to hyperglycemic animals significantly (P<0.05) decreased serum low density lipoprotein cholesterol (LDLC) concentration activity when compared with distilled water treated hyperglycemic animal. Administration of aqueous extract of *Sorghum bicolor and Pennisetum glaucum* at 750 and 1000mg/kg body weight to hyperglycemic animals significantly (P<0.05) decreased serum low density lipoprotein cholesterol (LDLC) concentration when compared with distilled water treated hyperglycemic animals (Table 5).

Table 5: Serum lipids of alloxan-induced hyperglycemic rats following oral administration of aqueous extract of *Sorghum bicolor* and *Pennisetum glaucum* seed

Treatment Group	Total Chol. (mmol/L)	Trig. (mmol/L)	HDLC (mmol/L)	LDLC (mmol/L)
A	1.82±0.04[a]	2.04±1.13[a]	15.42±2.06[d]	4.35±0.09[a]
B	10.04±0.72[c]	12.45±2.42[d]	3.41±0.13[a]	20.08±2.47[e]
C	2.20±0.24[a]	4.50±0.25[a]	11.42±1.41[c]	6.41±0.17[b]
D	3.45±0.17[a]	3.24±0.09[a]	9.04±0.52[c]	11.74±1.08[c]
E	2.25±0.45[a]	6.75±0.16[b]	10.56±1.06[c]	11.24±2.47[c]
F	6.54±1.24[b]	8.16±0.24[b]	6.72±0.21[b]	9.22±0.08[b]
G	7.42±0.72[b]	10.42±1.02[c]	9.77±0.24[c]	15.11±1.42[d]

Data are mean ± SEM of seven determinations. Test values with superscript different from their respective control down the column are significantly different (p<0.05)

A = Control; **B** = Diabetic rats + Distilled water; **C =** Diabetic rats + Glibenclamide; **D** = Diabetic rats + 750mg/kg body weight of *Sorghum bicolor* Extract; **E =** Diabetic rats + 1000 mg/kg body weight of *Sorghum bicolor* Extract; **F =** Diabetic rats + 750 mg/kg body weight of *Pennisetum glaucum* Extract; **G** = Diabetic rats + 1000 mg/kg body weight of *Pennisetum glaucum* Extract.

3.1.6 Histology of pancreas of alloxan-induced hyperglycemic rats following oral administration of aqueous extract of *Sorghum bicolor* and *Pennisetum glaucum* seed

Photomicrograph of a pancreas section stained by haematoxylin & eosin showing normal architecture, the parenchyma of the pancreas shows normal acinar cells (slender arrow) containing abundant granular eosinophilic cytoplasm, normal interlobular connective tissues (blue arrow) and septa (blue arrow) are seen. there normal islets of langerhans (white arrow) consisting of round to oval collections of endocrine cells (Plate 1a & b).

Photomicrograph of a pancreas section stained by haematoxylin & eosin showing damaged architecture, the parenchyma of the pancreas shows distorted acinar cells (slender arrow) containing reduced granular eosinophilic cytoplasm, deranged interlobular connective tissues (blue arrow) and septa (red arrow) are seen. There are a few damaged identifiable islets of Langerhans seen (Plate 2a & b).

Photomicrograph of a pancreas section stained by haematoxylin & eosin showing normal architecture, the parenchyma of the pancreas shows normal acinar cells (slender arrow)

containing abundant granular eosinophilic cytoplasm, normal interlobular connective tissues (blue arrow) and septa (red arrow) are seen. there normal islets of langerhans (white arrow) consisting of round to oval collections of endocrine cells (Plate 3a & b).

Photomicrograph of a pancreas section stained by haematoxylin & eosin showing normal architecture, the parenchyma of the pancreas shows normal acinar cells (slender arrow) containing abundant granular eosinophilic cytoplasm, normal interlobular connective tissues (blue arrow) and septa (red arrow) are seen. However, the islets of langerhans appear diffused with ill-defined borders (white arrow) (Plate 4a & b).

Photomicrograph of a pancreas section stained by haematoxylin & eosin showing attached lymphoid follicle (black arrow), the parenchyma of the pancreas shows normal acinar cells (slender arrow) containing abundant granular eosinophilic cytoplasm, normal interlobular connective tissues (red arrow) and septa (blue arrow) are seen. The islets of Langerhans seen show fluid accumulation (white arrow) (Plate 5a & b).

Photomicrograph of a pancreas section stained by haematoxylin & eosin showing moderate dilatation of vessels and congestion (black arrow), the parenchyma of the pancreas shows normal acinar cells (slender arrow) containing abundant granular eosinophilic cytoplasm, normal interlobular connective tissues (red arrow) and septa (bluearrow) are seen. There is moderately normal islet of langerhan seen (white arrow) (Plate 6a & b).

Photomicrograph of a pancreas section stained by haematoxylin & eosin showing normal architecture, the parenchyma of the pancreas shows normal acinar cells (slender arrow) containing abundant granular eosinophilic cytoplasm, normal interlobular connective tissues (red arrow) and septa (blue arrow) are seen. there islets of langerhans (white arrow) consisting of round to oval collections of endocrine cells (Plate 7a & b).

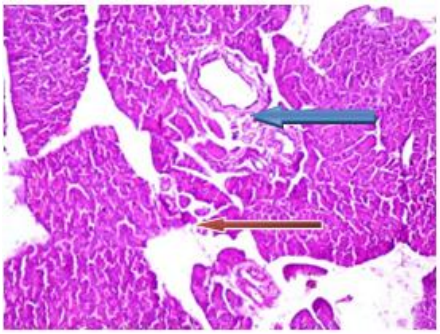

Plate 1a

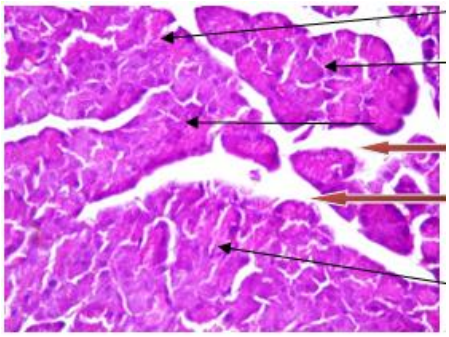

Plate 1b

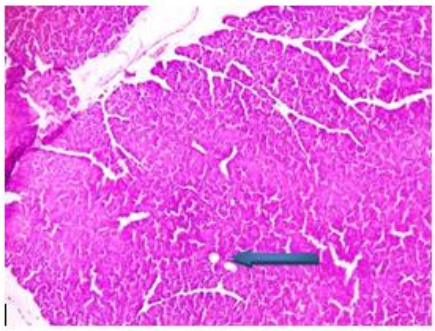

Plate 2a:

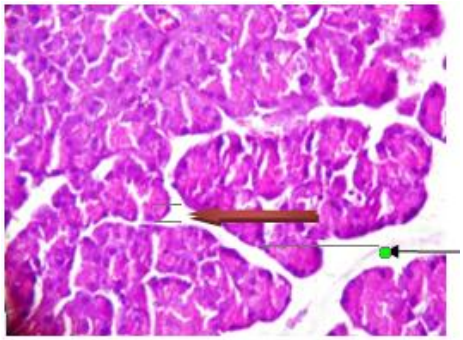

Plate 2b

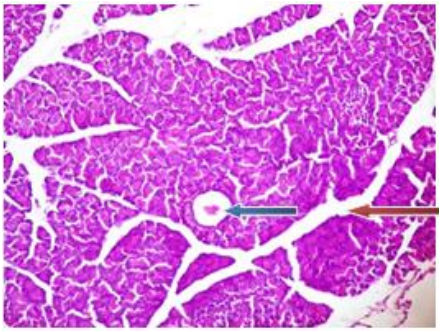

Plate 3a

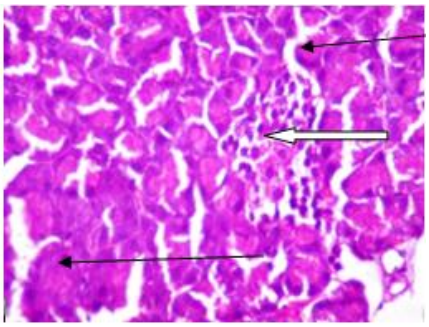

Plate 3b

Plate 1a: Photomicrograph of Pancreas of rats administered distilled water (X100 H&E)
Plate 1b: Photomicrograph of Pancreas of rats administered distilled water (X400 H&E)
Plate 2a: Photomicrograph of Pancreas of rats administered alloxan without treatment (X100 H&E)
Plate 2b: Photomicrograph of Pancreas of rats administered alloxan without treatment (X400 H&E)
Plate 3a: Photomicrograph of Pancreas of hyperglycemic rat treated with glibenclamide (X100 H&E)
Plate 3b: Photomicrograph of Pancreas of hyperglycemic rat treated with glibenclamide (X400 H&E

Plate 4a

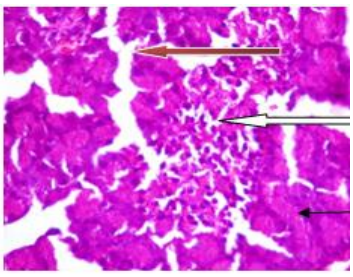

Plate 4b

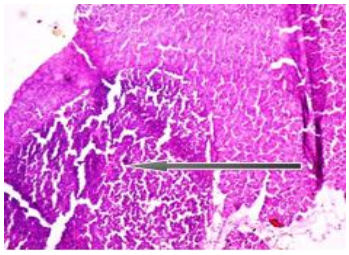

Plate 5a

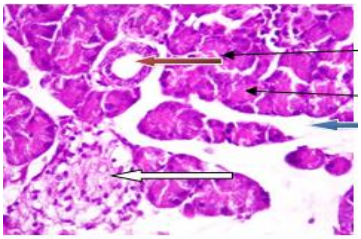

Plate 5b

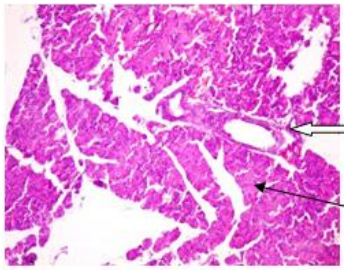

Plate 6a

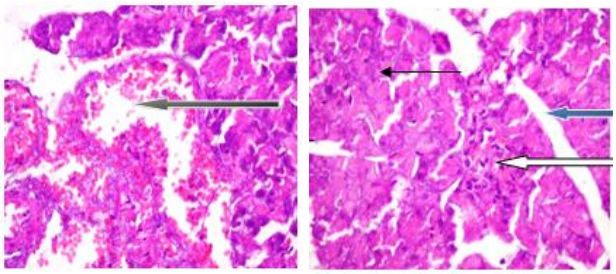

Plate 6b

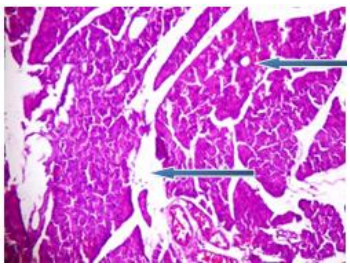

Plate 7a

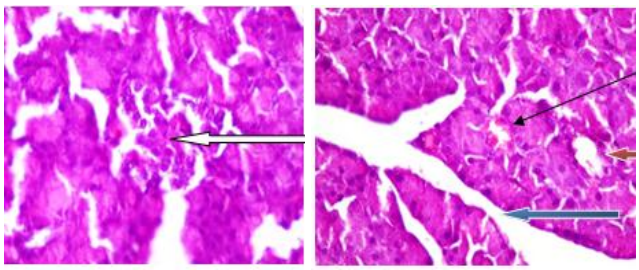

Plate 7b

Plate 4a: Photomicrograph of Pancreas of hyperglycemic rat treated with 750mg/kg body weight *Sorghum bicolor* (X100 H&E)

Plate 4b: Photomicrograph of Pancreas of hyperglycemic rat treated with 750mg/kg body weight *Sorghum bicolor* (X400 H&E)

Plate 5a: Photomicrograph of Pancreas of hyperglycemic rat treated with 1000mg/kg body weight *Sorghum bicolor* (X100 H&E)

Plate 5b: Photomicrograph of Pancreas of hyperglycemic rat treated with 1000mg/kg body weight *Sorghum bicolor* (X400 H&E)

Plate 6a: Photomicrograph of Pancreas of hyperglycemic rat treated with 750mg/kg body weight *Pennisetum glaucum* (X100 H&E)

Plate 6b: Photomicrograph of Pancreas of hyperglycemic rat treated with 750mg/kg body weight *Pennisetum glaucum* (X400 H&E)

Plate 7a: Photomicrograph of Pancreas of hyperglycemic rat treated with 1000mg/kg body weight *Pennisetum glaucum* (X100 H&E)

Plate 7b: Photomicrograph of Pancreas of hyperglycemic rat treated with 1000mg/kg body weight *Pennisetum glaucum* (X400 H&E)

3.2 Discussion

3.2.1 Blood glucose

Persistent hyperglycemia is the hallmark of diabetes mellitus and represents a major risk factor for the development of metabolic complications associated with increased morbidity and mortality among diabetic individuals (American Diabetes Association [ADA], 2024). Experimental induction of diabetes using alloxan is widely employed in biomedical research due to its selective cytotoxic effects on pancreatic β-cells of the islets of Langerhans. Alloxan induces diabetes primarily through the generation of reactive oxygen species (ROS), leading to β-cell destruction, impaired insulin secretion, and consequent elevation of blood glucose levels (Lenzen, 2008). The resulting insulin deficiency compromises glucose uptake and utilization by peripheral tissues, thereby promoting hyperglycemia and associated metabolic disturbances such as dyslipidemia, protein catabolism, and oxidative stress (Szkudelski, 2012). Assessment of blood glucose concentration in alloxan-induced diabetic experimental animals therefore serves as an important quantitative indicator for evaluating the severity of diabetes and the efficacy of antidiabetic interventions. In the management of diabetes mellitus, oral hypoglycemic agents such as Glibenclamide are commonly used as standard therapeutic agents. Glibenclamide, a second-generation sulfonylurea, exerts its hypoglycemic effect primarily by stimulating insulin release from functional pancreatic β-cells through inhibition of ATP-sensitive potassium channels, thereby enhancing glucose utilization and reducing circulating blood glucose levels (Rena *et al*., 2017).

The observed hypoglycemic effects of sorghum and millet extracts in experimental studies may be attributed to their high content of bioactive phytochemicals, particularly polyphenols, tannins, and dietary fiber. Previous investigations have demonstrated that sorghum extracts rich in condensed tannins significantly reduce serum glucose concentrations in streptozotocin-induced diabetic animal models and high-fat diet-induced metabolic disorders (Park *et al*., 2012). Sorghum polyphenols have also been shown to inhibit the activities of key carbohydrate-hydrolyzing enzymes such as α-amylase and α-glucosidase, which are important therapeutic targets in diabetes management because they delay carbohydrate digestion and glucose absorption in the intestine (Kim *et al*., 2011). Furthermore, consumption of sorghum-based diets has been associated with reductions in fasting blood glucose and improved glycemic responses in individuals with type 2 diabetes mellitus, effects partly attributed to the high dietary fiber content of sorghum grains (Lakshmi *et al*., 1996).

Dietary fiber contributes to glycemic regulation by delaying gastric emptying, prolonging intestinal transit time, and reducing the rate of carbohydrate absorption, ultimately leading to improved postprandial glucose control (Ray *et al*., 1983). Similar antihyperglycemic effects have also been reported for other polyphenol-rich plant extracts, including cocoa and *Salacia reticulata*, which demonstrated significant reductions in serum glucose levels in diabetic animal models through antioxidant and insulin-sensitizing mechanisms (Ruzaidi *et al*., 2005; Akase *et al*., 2011).

Likewise, Pearl millet (*Pennisetum glaucum*) has been reported to possess significant blood glucose-lowering properties owing to its abundance of phenolic compounds, dietary fiber, and

micronutrients. The hypoglycemic effects of pearl millet may involve stimulation of pancreatic insulin secretion, enhancement of insulin sensitivity in peripheral tissues, inhibition of intestinal glucose absorption, and attenuation of oxidative stress associated with diabetes mellitus (Anitha *et al*., 2017). Collectively, these findings suggest that the antidiabetic activities of sorghum and pearl millet may be mediated through multiple complementary mechanisms that improve glucose homeostasis and metabolic regulation.

3.2.2 Serum antioxidant status

Hyperglycemia-associated glucose toxicity in diabetes mellitus is known to enhance the production of reactive oxygen species (ROS), thereby promoting oxidative stress and disrupting cellular antioxidant defense mechanisms (Maritim *et al*., 2003). Excessive ROS generation contributes significantly to the pathogenesis of diabetic complications through oxidative damage to lipids, proteins, and nucleic acids. In response to increased oxidative stress, both enzymatic and non-enzymatic antioxidant defense systems are often activated; however, prolonged oxidative imbalance may overwhelm these protective mechanisms, resulting in cellular dysfunction and tissue injury (Asmat *et al*., 2016).

In the present context, treatment with sorghum and millet extracts appeared to normalize ROS levels and improve antioxidant status when compared with untreated diabetic controls, which exhibited elevated oxidative stress markers and altered antioxidant enzyme activities. These observations are consistent with previous reports demonstrating significant reductions in lipid peroxidation products and oxidative stress biomarkers in diabetic experimental animals treated with natural antioxidant-rich substances such as camel milk and *Ginkgo biloba* extract (Hassan & Emam, 2005). The antioxidant effects of these plant-derived interventions may therefore contribute to their protective role against diabetes-induced oxidative damage.

The antioxidant potential of sorghum and pearl millet is largely attributed to their abundance of phenolic compounds, including flavonoids, tannins, and anthocyanins. Phenolic compounds function primarily as reducing agents, hydrogen donors, and singlet oxygen quenchers, thereby neutralizing free radicals and inhibiting oxidative chain reactions (Kang *et al*., 2006). The high free radical scavenging activity observed in sorghum and millet extracts may therefore be mediated by these bioactive phytochemicals. Several studies have demonstrated that cereal polyphenols possess potent antioxidant activities capable of attenuating oxidative stress-related metabolic disorders, including diabetes mellitus (Sharma *et al*., 2021).

Despite the established antioxidant potential of phenolic compounds, assessment of antioxidant activity using single in vitro assays may produce variable or underestimated results. For instance, some phenolic compounds react slowly with 2,2-diphenyl-1-picrylhydrazyl (DPPH), reaching reaction equilibrium only after prolonged incubation periods (Bondet *et al*., 1997). Furthermore, interference from naturally pigmented compounds such as anthocyanins may affect spectrophotometric measurements and lead to underestimation of antioxidant capacity in DPPH assays (Arnao, 2000). Consequently, it has been recommended that multiple analytical methods be employed when evaluating antioxidant activities of plant extracts due to differences in assay sensitivity, reaction mechanisms, and specificity toward various antioxidant compounds (Schlesier *et al*., 2002). The use of complementary antioxidant assays

therefore provides a more comprehensive evaluation of the free radical scavenging properties of sorghum and millet phytochemicals.

3.2.3 Liver function indices

Alloxan-induced diabetes is associated with severe pancreatic β-cell destruction, resulting in impaired insulin secretion, persistent hyperglycemia, and metabolic disturbances that contribute to hepatic dysfunction (Lenzen, 2008). In addition to its diabetogenic effects, alloxan promotes oxidative stress and inflammatory responses that may compromise liver integrity and function. Hepatic injury in diabetic conditions is commonly reflected by elevated serum activities of liver enzymes such as alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP), which serve as important biochemical indicators of hepatocellular damage and impaired liver function (Sakurai *et al*., 2001).

The reduction in serum ALT, AST, and ALP activities observed following treatment with sorghum and millet extracts suggests a significant improvement in hepatic function and structural integrity. These findings indicate that the extracts may possess hepatoprotective properties capable of attenuating diabetes-induced liver injury. The hepatoprotective effects observed in this study may largely be attributed to the antioxidant activities of the phytochemical constituents present in sorghum and pearl millet, particularly polyphenols, flavonoids, tannins, and anthocyanins, which are known to neutralize reactive oxygen species and reduce oxidative damage in hepatic tissues (Awika & Rooney, 2004).

Oxidative stress plays a crucial role in the progression of diabetic liver injury through lipid peroxidation, mitochondrial dysfunction, and inflammatory signaling pathways. Therefore, antioxidant-rich plant extracts may help restore hepatic antioxidant defense systems and preserve cellular membrane integrity (Asmat *et al*., 2016). The current findings are consistent with previous studies reporting hepatoprotective and antioxidant activities of phytochemical-rich fermented products such as red ginseng and mung bean extracts in diabetic experimental models (Ali *et al*., 2013; Yeap *et al*., 2015). These studies demonstrated significant reductions in liver enzyme activities and improvements in hepatic histoarchitecture following treatment with natural antioxidant compounds.

Furthermore, the hepatoprotective potential of sorghum and millet may also be linked to their ability to improve glycemic control and reduce hyperglycemia-induced oxidative stress, thereby limiting secondary hepatic complications associated with diabetes mellitus. Collectively, these findings support the therapeutic potential of sorghum and pearl millet extracts as natural agents for the management of diabetes-related hepatic dysfunction.

3.2 4 Kidney function indices

Elevated serum concentrations of urea, uric acid, and creatinine are common biochemical manifestations of diabetes mellitus and are widely recognized indicators of renal dysfunction and impaired metabolic homeostasis in diabetic conditions (Resmi *et al*., 2001; Ezeigbo & Asuzu, 2012). Chronic hyperglycemia and oxidative stress associated with diabetes contribute significantly to alterations in protein metabolism, renal filtration, and nitrogenous waste excretion, thereby leading to accumulation of these metabolites in the bloodstream (Forbes &

Cooper, 2013). Increased serum urea levels observed in untreated diabetic animals may be attributed to enhanced protein catabolism and accelerated amino acid degradation resulting from insulin deficiency. Under diabetic conditions, impaired glucose utilization promotes proteolysis and gluconeogenesis, ultimately increasing hepatic urea production through activation of the urea cycle.

Furthermore, disturbances in carbohydrate metabolism associated with diabetes can alter glycolytic pathways and redirect glucose metabolism toward the pentose phosphate pathway, resulting in increased production of phosphoribosyl pyrophosphate (PRPP), a key intermediate involved in purine metabolism and uric acid synthesis (Lal *et al*., 2009). Consequently, elevated serum uric acid concentrations frequently occur in diabetic conditions and may contribute to oxidative stress, endothelial dysfunction, and progression of diabetic nephropathy (Johnson *et al*., 2013).

Similarly, hypercreatininemia observed in untreated alloxan-induced diabetic animals may indicate impaired renal clearance and compromised kidney function. Elevated creatinine levels are commonly associated with reduced glomerular filtration rate and renal tissue damage resulting from persistent hyperglycemia and oxidative stress (Sokeng *et al*., 2005). Renal dysfunction remains one of the most serious complications of diabetes mellitus and is characterized by progressive deterioration of kidney function and accumulation of metabolic waste products in circulation.

Treatment with aqueous extracts of Sorghum bicolor and Pearl millet (*Pennisetum glaucum*) at doses of 750 and 1000 mg/kg body weight significantly ameliorated the elevated levels of urea, uric acid, and creatinine in alloxan-induced diabetic animals. The observed reductions in these biochemical markers suggest possible nephroprotective and metabolic regulatory effects of the extracts. These protective effects may be attributed to the antioxidant and antihyperglycemic properties of their phytochemical constituents, particularly polyphenols, flavonoids, tannins, and dietary fiber, which are capable of reducing oxidative stress, improving glucose metabolism, and preserving renal integrity (Awika & Rooney, 2004; Sharma *et al*., 2021).

The ability of sorghum and millet extracts to restore altered renal biomarkers further supports their therapeutic potential in the management of diabetes-associated metabolic complications. Therefore, these findings suggest that aqueous extracts of *Sorghum bicolor* and *Pennisetum glaucum* may serve as promising natural interventions for mitigating renal dysfunction and other metabolic abnormalities associated with diabetes mellitus.

3.2.5 Serum lipids

Alloxan-induced diabetes mellitus is frequently associated with significant alterations in lipid metabolism, leading to elevated serum levels of total cholesterol, triglycerides, low-density lipoprotein cholesterol (LDL-C), and very low-density lipoprotein cholesterol (VLDL-C), a condition commonly referred to as diabetic dyslipidemia (Goldberg, 2001). Hypercholesterolemia observed in diabetic conditions is largely attributed to impaired insulin activity, which disrupts lipid homeostasis and enhances hepatic cholesterogenesis. Persistent elevation of circulating cholesterol contributes to the development of atherosclerosis through deposition of lipids within arterial walls, thereby increasing the risk of cardiovascular diseases,

stroke, and other vascular complications associated with diabetes mellitus (Granner *et al*., 1996).

Under diabetic conditions, serum high-density lipoprotein cholesterol (HDL-C), commonly regarded as "good cholesterol," is typically reduced, whereas levels of triglycerides, LDL-C, and VLDL-C are elevated (Girard-Mauduit, 2010). HDL-C plays a protective role in lipid metabolism by facilitating reverse cholesterol transport from peripheral tissues to the liver for metabolism and excretion. Conversely, LDL-C and VLDL-C transport cholesterol from the liver to peripheral tissues, and excessive accumulation of these lipoproteins contributes significantly to plaque formation and vascular dysfunction (Lyons, 1992). Consequently, diabetic dyslipidemia represents a major risk factor for coronary heart disease and other cardiovascular complications frequently observed in diabetic patients (Daniel *et al*., 2011).

In the present study, administration of aqueous extracts of Sorghum bicolor and Pearl millet (*Pennisetum glaucum*) significantly reduced serum concentrations of triglycerides, total cholesterol, and LDL-C in alloxan-induced diabetic animals. These findings suggest that the extracts possess hypolipidemic properties capable of improving lipid metabolism and reducing cardiovascular risk associated with diabetes mellitus. Previous investigations have similarly demonstrated that sorghum and millet-derived phytochemicals exert cholesterol-lowering effects by suppressing hepatic cholesterol synthesis and enhancing fecal cholesterol excretion (He *et al*., 2007; Kim *et al*., 2010).

Furthermore, sorghum extracts rich in polyphenols and tannins have been reported to significantly decrease serum total cholesterol concentrations, potentially through modulation of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase activity, the rate-limiting enzyme involved in cholesterol biosynthesis (Chung *et al*., 2011). The hypolipidemic effects of sorghum and millet may also be linked to their high dietary fiber content, antioxidant activities, and ability to improve insulin sensitivity, all of which contribute to improved lipid utilization and reduced lipid accumulation in tissues (Awika & Rooney, 2004).

Additionally, the antioxidant properties of the phytochemical constituents present in sorghum and millet may protect against lipid peroxidation and oxidative modification of LDL particles, processes that play critical roles in the pathogenesis of atherosclerosis and cardiovascular disease (Asmat *et al*., 2016). Therefore, the observed improvements in lipid profiles following treatment with sorghum and millet extracts indicate their potential therapeutic value in the management of diabetic dyslipidemia and associated cardiovascular complications. However, further in vivo and mechanistic studies are required to elucidate the precise molecular pathways involved, particularly the role of HMG-CoA reductase inhibition in mediating the cholesterol-lowering effects of these extracts.

3.2.6 Histological studies

Induction of hyperglycemia by alloxan in experimental animals is known to produce severe cytotoxic effects on pancreatic β-cells, leading to structural and functional impairment of the pancreas. The diabetogenic action of alloxan is primarily mediated through selective accumulation within pancreatic β-cells via the glucose transporter GLUT2, followed by the generation of reactive oxygen species (ROS), including superoxide radicals, hydrogen peroxide, and hydroxyl radicals, which collectively induce oxidative stress and cellular damage

(Lenzen, 2008). One proposed mechanism underlying alloxan-induced β-cell destruction involves inhibition of endogenous antioxidant defense enzymes, particularly superoxide dismutase (SOD), thereby reducing the capacity of pancreatic cells to neutralize oxidative stress (Grankvist *et al*., 1981).

In the present study, the marked damage observed in the histoarchitecture of pancreatic tissues from untreated diabetic animals suggests extensive β-cell degeneration resulting from oxidative injury and compromised antioxidant defense mechanisms. Reduced SOD activity may have contributed significantly to the inability of pancreatic tissues to counteract ROS accumulation, leading to redox imbalance, lipid peroxidation, and eventual β-cell dysfunction. These pathological changes are consistent with previous reports demonstrating pancreatic necrosis, islet shrinkage, and loss of β-cell integrity following alloxan administration (Lenzen, 2008).

Treatment with aqueous extracts of Sorghum bicolor and Pearl millet (*Pennisetum glaucum*) at doses of 750 and 1000 mg/kg body weight, as well as the standard antidiabetic drug Glibenclamide, significantly ameliorated the pancreatic damage induced by alloxan. Histopathological improvements observed in treated groups suggest that both extracts may possess protective effects on pancreatic β-cells, likely through attenuation of oxidative stress and preservation of cellular integrity. The restoration of pancreatic architecture may indicate improved β-cell survival and enhanced insulin secretory function.

The protective effects of sorghum and millet extracts may be attributed to their rich phytochemical composition, particularly polyphenols, flavonoids, tannins, and anthocyanins, which possess potent antioxidant and free radical-scavenging activities (Awika & Rooney, 2004). These bioactive compounds may enhance endogenous antioxidant defenses, reduce oxidative damage, and stabilize cellular membranes, thereby preserving pancreatic tissue integrity (Sharma *et al*., 2021). Additionally, reduction in oxidative stress may promote regeneration or recovery of partially damaged β-cells, contributing to improved glycemic control.

These findings are consistent with earlier studies demonstrating that antioxidant-rich plant extracts can protect pancreatic β-cells from oxidative injury and improve pancreatic histology in diabetic animal models (Eroschenko, 2000). Therefore, the aqueous extracts of *Sorghum bicolor* and *Pennisetum glaucum* may exert their antidiabetic effects, at least in part, through pancreatic cytoprotection, reduction of oxidative stress, and preservation of β-cell functional integrity.

4.0 CONCLUSION AND RECOMMENDATIONS

4.1 Conclusion

The findings of this study demonstrate that aqueous extracts of Sorghum bicolor and Pearl millet (*Pennisetum glaucum*) possess significant antidiabetic properties in alloxan-induced diabetic experimental animals. Administration of the extracts resulted in marked reductions in blood glucose levels, improvement in lipid profile parameters, restoration of liver and kidney function biomarkers, attenuation of oxidative stress, and preservation of pancreatic histoarchitecture. Among the treatment groups, pearl millet extract administered at 1000 mg/kg body weight exhibited the most pronounced antidiabetic effect.

The extracts also demonstrated protective effects against diabetes-induced histological alterations and biochemical abnormalities, suggesting their potential therapeutic value in the management of diabetes mellitus and its associated complications. The observed beneficial effects may be attributed to the rich phytochemical composition of sorghum and millet, particularly their phenolic compounds, flavonoids, tannins, anthocyanins, and dietary fiber, which possess antioxidant, antihyperglycemic, hypolipidemic, and cytoprotective properties.

Based on the findings of this study, the possible mechanisms through which the extracts exert their antidiabetic effects may include:

1. Facilitation of pancreatic insulin secretion through protection and preservation of pancreatic β-cell integrity.
2. Enhancement of glucose uptake and utilization by peripheral tissues.
3. Improvement of insulin sensitivity and modulation of glucose metabolism.
4. Reduction of oxidative stress through free radical scavenging and enhancement of antioxidant defense systems.
5. Regulation of lipid metabolism and reduction of diabetes-associated dyslipidemia.

Overall, the results suggest that aqueous extracts of *Sorghum bicolor* and *Pennisetum glaucum* may serve as promising natural therapeutic agents for the management of diabetes mellitus and its metabolic complications.

4.2 Recommendations

Based on the outcomes of this study, the following recommendations are proposed:

1. Further studies should be conducted to isolate, characterize, and identify the specific bioactive compounds responsible for the antidiabetic activities of *Sorghum bicolor* and *Pennisetum glaucum*.
2. Detailed molecular and mechanistic investigations are necessary to elucidate the precise pathways involved in the antihyperglycemic and antioxidant effects of the extracts.
3. Long-term toxicity and safety studies should be performed to establish the safety profile of the extracts for prolonged therapeutic use.
4. Clinical trials involving human subjects are recommended to validate the efficacy and safety of sorghum and millet extracts in the management of diabetes mellitus.
5. Further research should evaluate the effects of the extracts on other diabetes-related complications such as nephropathy, neuropathy, cardiovascular dysfunction, and retinopathy.
6. Development of functional foods or nutraceutical formulations incorporating sorghum and millet may provide affordable dietary interventions for the prevention and

management of diabetes, particularly in low- and middle-income countries where the burden of the disease is rapidly increasing.

Ethical Approval Statement

All experimental procedures involving animals were conducted in accordance with institutional guidelines for the care and use of laboratory animals and complied with internationally accepted ethical standards for animal experimentation.

Conflict of Interest Statement

The authors declare no conflict of interest.

Funding Statement

This research received no external funding.

Data Availability Statement

The datasets generated and analyzed during the current study are available from the corresponding author upon reasonable request.

Author Contributions

Conceptualization, methodology, investigation, data analysis, and manuscript preparation were performed by the authors.

REFERENCES

Aderibigbe, O. R., Ezekiel, O. O., Owolade, S. O., Korese, J. K., & Sturm, B. (2021). Exploring the potentials of underutilized grain amaranth, sorghum, and millet in combating food insecurity and malnutrition in Africa. *Frontiers in Sustainable Food Systems, 5*, 1–15. https://doi.org/10.3389/fsufs.2021.XXXXXX

Akase, T., Shimada, T., Harasawa, Y., Ikeya, Y., Nagai, E., & Iizuka, S. (2011). Preventive effects of *Salacia reticulata* on obesity and metabolic disorders in TSOD mice. *Evidence-Based Complementary and Alternative Medicine, 2011*, 484590. https://doi.org/10.1155/2011/484590

Ali, N. M., Yeap, S. K., Ho, W. Y., Beh, B. K., Tan, S. W., Tan, S. G., & Alitheen, N. B. (2013). The promising future of chia (*Salvia hispanica* L.) as a functional food for human health. *Journal of Biomedicine and Biotechnology, 2013*, 171956. https://doi.org/10.1155/2013/171956

American Diabetes Association. (2024). Standards of care in diabetes—2024. *Diabetes Care, 47*(Supplement_1), S1–S350. https://doi.org/10.2337/dc24-SINT

Anitha, S., Kane-Potaka, J., Tsusaka, T. W., Botha, R., & Rajendran, A. (2017). Assessing the role of millets in combating nutritional insecurity and hidden hunger: A review. *Frontiers in Nutrition, 4*, 10. https://doi.org/10.3389/fnut.2017.00010

Anitha, S., Kane-Potaka, J., Tsusaka, T. W., Botha, R., & Rajendran, A. (2017). Millets can improve iron status and reduce iron deficiency anemia: A systematic review and meta-analysis. *Frontiers in Nutrition*, *4*, 56. https://doi.org/10.3389/fnut.2017.00056

Arnao, M. B. (2000). Some methodological problems in antioxidant activity determination using chromogen radicals. *Trends in Food Science & Technology*, *11*(11), 419–421.

Asmat, U., Abad, K., & Ismail, K. (2016). Diabetes mellitus and oxidative stress: A concise review. *Saudi Pharmaceutical Journal*, *24*(5), 547–553. https://doi.org/10.1016/j.jsps.2015.03.013

Awika, J. M., & Rooney, L. W. (2004). Sorghum phytochemicals and their potential impact on human health. *Phytochemistry*, *65*(9), 1199–1221. https://doi.org/10.1016/j.phytochem.2004.04.001

Bathaie, S. Z., & Mousavi, S. Z. (2018). Dietary phytochemicals in diabetes management: A review. *EXCLI Journal*, *17*, 64–104. https://doi.org/10.17179/excli2017-999

Beers, R. F., & Sizer, I. W. (1952). A spectrophotometric method for catalase activity. *Journal of Biological Chemistry*, *195*(1), 133–140.

Bondet, V., Brand-Williams, W., & Berset, C. (1997). Kinetics of antioxidant activity using DPPH method. *LWT – Food Science and Technology*, *30*(6), 609–615.

Buege, J. A., & Aust, S. D. (1978). Microsomal lipid peroxidation. *Methods in Enzymology*, *52*, 302–310. https://doi.org/10.1016/S0076-6879(78)52032-6

Chung, I. M., Kim, E. H., Yeo, M. A., Kim, S. J., Seo, M. C., & Moon, H. I. (2011). Antidiabetic effects of sorghum bran extracts. *Food Chemistry*, *124*(3), 1053–1058.

Daniel, R. S., Devi, K. S., Augusti, K. T., & Nair, C. R. S. (2011). Mechanism of flavonoids in antidiabetic activity. *Indian Journal of Experimental Biology*, *41*(4), 296–303.

Dillon, S. L., Shapter, F. M., Henry, R. J., Cordeiro, G., Izquierdo, L., & Lee, L. S. (2009). Domestication and genetic resources of sorghum. *Annals of Botany*, *100*(5), 975–989. https://doi.org/10.1093/aob/mcm192

Ezeigbo, I. I., & Asuzu, I. U. (2012). Antidiabetic activity of *Vernonia amygdalina* in alloxan-induced rats. *African Journal of Biotechnology*, *11*(8), 2081–2087.

FAO. (2022). FAOSTAT statistical database. https://www.fao.org/faostat

FAO. (2022). Sorghum and millets in human nutrition. Food and Agriculture Organization.

FAO. (2023). FAOSTAT statistical database. https://www.fao.org/faostat

Forbes, J. M., & Cooper, M. E. (2013). Mechanisms of diabetic complications. *Physiological Reviews*, *93*(1), 137–188.

Fredrickson, D. S., Levy, R. I., & Lees, R. S. (1967). Lipoprotein metabolism. *New England Journal of Medicine*, *276*, 148–156.

Friedewald, W. T., Levy, R. I., & Fredrickson, D. S. (1972). LDL estimation formula. *Clinical Chemistry*, *18*(6), 499–502. https://doi.org/10.1093/clinchem/18.6.499

GBD 2021 Diabetes Collaborators. (2023). Global burden of diabetes. *The Lancet*, *402*(10397), 203–234. https://doi.org/10.1016/S0140-6736(23)01301-6

Geijselaers, S. L. C., et al. (2017). Insulin resistance and diabetes risk. *Diabetologia*, *60*(4), 568–578.

Girard-Mauduit, C. (2010). Dyslipidemia in diabetes. *Diabetes & Metabolism*, *36*(6), 419–426.

Goldberg, I. J. (2001). Diabetic dyslipidemia. *The Journal of Clinical Endocrinology & Metabolism*, *86*(3), 965–971.

Grankvist, K., Marklund, S. L., & Täljedal, I. B. (1981). Superoxide dismutase protection in diabetes. *Nature*, *294*, 158–160.

Granner, D. K., Mayes, P. A., & Murray, R. K. (1996). *Harper's biochemistry* (24th ed.). Appleton & Lange.

Hainline, A., Cooper, G. R., & Jung, D. H. (1980). Clinical chemistry manual. AACC.

Hassan, N. S., & Emam, M. A. (2005). Protective effects against alloxan diabetes. *Journal of Applied Sciences Research*, *1*(2), 108–114.

He, J., Guo, H., & Wang, Y. (2007). Hypocholesterolemic effects of sorghum. *Journal of Nutritional Biochemistry*, *18*(6), 408–414.

IDF. (2021). *Diabetes Atlas* (10th ed.). International Diabetes Federation.

Jaffe, M. (1941). Creatinine reaction. *Zeitschrift für Physiologische Chemie*, *10*, 391–400.

Johnson, R. J., et al. (2013). Sugar, uric acid, and diabetes. *Diabetes*, *62*(10), 3307–3315.

Kim, J. S., Hyun, T. K., & Kim, M. J. (2010). Sorghum antioxidant effects. *Food Chemistry*, *115*(4), 1236–1241.

Lenzen, S. (2008). Alloxan and streptozotocin diabetes. *Diabetologia*, *51*(2), 216–226.

Maritim, A. C., Sanders, R. A., & Watkins, J. B. (2003). Oxidative stress in diabetes. *Journal of Biochemical and Molecular Toxicology*, *17*(1), 24–38.

Misra, H. P., & Fridovich, I. (1972). Superoxide dismutase assay. *Journal of Biological Chemistry*, *247*(10), 3170–3175.

Reitman, S., & Frankel, S. (1957). AST/ALT assay. *American Journal of Clinical Pathology*, *28*(1), 56–63. https://doi.org/10.1093/ajcp/28.1.56

Rosenthal, H. L. (1955). Urea determination. *Analytical Chemistry*, *27*(6), 1272–1274.

Szkudelski, T. (2012). Streptozotocin model of diabetes. *Experimental Biology and Medicine*, *237*(5), 481–490.

Tietz, N. W. (1990). *Clinical guide to laboratory tests* (2nd ed.). W. B. Saunders.

Tietz, N. W. (1995). *Clinical guide to laboratory tests* (3rd ed.). W. B. Saunders.

WHO. (2023). Diabetes fact sheet. https://www.who.int

Wright, P. J., Leathwood, P. D., & Plummer, D. T. (1972). Liver enzymes study. *Enzymologia*, *42*, 317–327.