Trop J Nat Prod Res, September 2024; 8(9): 8424-8431 ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

Tropical Journal of Natural Product Research

Available online at https://www.tjnpr.org

Original Research Article

Hypoglycaemic and Histophatological Effect of *Myrianthus arboreus* in Alloxan-Induced Diabetic Rats

Joseph O. Ezugworie[1], Agatha O. Anene[1], Fredrick O. Ugbede[2]*

[1]*Department of Anatomy, University of Nigeria, Enugu Campus, Nigeria*
[2]*Department of Physics, Dennis Osadebay University, Asaba, Nigeria*

ARTICLE INFO

Article history:
Received: 11 August 2024
Revised: 24 August 2024
Accepted: 10 September 2024
Published online: 01 October 2024

ABSTRACT

Diabetes mellitus is a common global health disorder characterized by chronic hyperglycaemia owing to decreased insulin secretion or sensitivity. Medicinal plants have continued to gain more relevance in diabetes treatment owing to their reliance as alternative therapy. This study examined the histological effects of *Myrianthus arboreus* leaf extracts on the pancreas of alloxan-induced diabetic albino Wister rats. Two extracts of *Myrianthus arboreus* leaves were formulated; aqueous extract (AEME) and ethanol extract (EEMA). Qualitative and quantitative phytochemical assays were investigated on both extracts. Induction of diabetes was made after overnight fasting of rats by a single intraperitoneal injection of 150 mg/kg (body weight) solution of alloxan. AEMA and EEMA were administered at 100, 200, and 400 mg/kg body weight daily for 28 days. Also, glibenclamide was administered at 5 mg/kg body weight to the positive control group. The rats were monitored every week for body weight and blood glucose levels. They were fasted 12 hours after the last treatment before sacrifice by cervical decapitation under ether anesthesia. A histological study was carried out on pancreas tissues. All treated groups, except the untreated group, had significant ($p < 0.05$) weight gain. Administration of AEMA and EEMA led to a significant ($p < 0.05$) decrease in blood glucose level, restoration of islets of Langerhans, and increased pancreatic beta cells compared to the diabetic group. Therefore, *Myrianthus arboreus* leaves showed potent antidiabetic properties, as evidenced by the increase in several beta cells and a decrease in blood glucose levels.

Keywords: Diabetes mellitus, Pancreas, Leaf extract, *Myrianthus arboreus*.

Introduction

Diabetes mellitus (DM) is a common global health disease with over 463 million prevalence cases worldwide,[1,2] and one of the leading causes of death.[3] According to the World Health Organization (WHO), over 80% of death counts from DM majorly occur in low to medium-income countries.[4] DM has already been marked ahead to be the 7th leading cause of death worldwide by the year 2030.[5]DM is a metabolic disorder with features of chronic hyperglycaemia with complete or comparative deficiencies in insulin production, sensitivity, and secretion.[6] The two common effects of DM are decreased insulin secretion due to the destruction of insulin-secreting beta cells of the pancreas and insulin insensitivity by the peripheral tissues for glucose metabolism. These two effects result in an unusually high blood glucose levels, referred to as hyperglycemia. Progressive complications of diabetes result from increased oxidative stress brought about by the increase in free radical production and associated imbalance between antioxidant defense systems.[7,8]

Several complications of DM include nephropathy, cardiomyopathy, and longstanding impairments such as retinopathy, stroke, neural damage, and delayed wound healing.[8]Treatment of DM involves the use of conventional antidiabetic drugs, insulin injections, and dietary lifestyle modification, with each having its drawbacks. Patient inconsistency in lifestyle dietary adjustment and the anxiety of insulin injection are major setbacks to their uses. Also, conventional antidiabetic drugs are characterized by undesirable side effects, which include gastrointestinal disorders, weight gain, hypersensitivity, nausea, diarrhea, and liver and heart failure.[1,9] Aside from this profile of undesirable effects associated with antidiabetic drugs, there is also the issue of affordability occasioned by their high prices, especially in developing regions. These shortfalls associated with commercially available synthesized drugs have increased the reliance on alternative therapy for the management of DM. Medicinal plants have been deployed as alternative treatment and management plans for diabetes mellitus in many countries worldwide. In recent times, the WHO has encouraged scientific expansion of the frontiers of hypoglycemic properties of various plantswith possible recommendations for their use in the management of DM.[10] So far, several medicinal plant parts and products have been studied to possess antidiabetic properties essential for improving glucose metabolism and total well-being conditions of individuals with diabetes.[9,11] Medicinal plants possess ample antioxidants and phytochemicals such as glycosides, phenolic compounds, alkaloids, terpenoids, flavonoids, and coumarins to control glucose metabolism. Clinical evidence has maintained the potencies and efficacies of medicinal plants in glucose homeostasis.[12]*Myrianthus arboreus*is a forest tree common to the West, East, and Central African regions. It occurs in the rainforest, semi-deciduous, and swamp forests.[13] It is a native wild plant of the urticaceae family,[14] and commonly used in foods and herbs.[13] The roots, stem bark and leaf extracts of the plant are used in traditional medical practices to manage various illnesses and disorders.[13] Reported studies showed that *Myrianthus aboreus* has an affluence of bioactive compounds and phytochemicals, such as saponins, alkaloids, tannins, flavonoids, myriantic acid, triterpenes, pentacyclic triterpenic diacids.[15,16,17] The plant extracts have been indicated for antibacterial activity[18]antinociceptive effects,[13] exhibit antioxidants,[19] and antidiabetic potential.[20] Although the antidiabetic properties of *Myrianthus aboreus*

*Corresponding author. Email: fredrick.ugbede@dou.edu.ng
Tel: + 08135535697

Citation: Ezugworie JO, Anene AO, Ugbede FO. Hypoglycaemic and Histophatological Effect of *Myrianthus arboreus* in Alloxan-Induced Diabetic Rats. Trop J Nat Prod Res. 2024; 8(9): 8424 – 8431. https://doi.org/10.26538/tjnpr/v8i9.26

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

stem bark and roots have been scientifically studied using a rat model and used in traditional diabetes management, there is still a paucity of scientific knowledge of its leaves extract in diabetes management. This study investigated the histological changes of aqueous and ethanol leaf extracts of *Myrianthus aboreus* in alloxan-induced diabetic Wister rats.

Materials and Methods

Chemicals

Alloxan monohydrate was obtained from Sigma-Aldrich Chemical Co. (USA). Glibenclamide was acquired from Wellona Pharma (India). All the chemicals and reagents were of the analytical grade and commercially available.

Plant collection and identification

Young fresh leaves of *Myrianthus arboreus* were obtained from a farm in Issele-Uku village in Delta state, Nigeria, in the first week of November 2019. The leaves were identified and authenticated by a plant taxonomist/technologist (Mr. C. J. Onyeukwu) at Plant Science and Biotechnology department, University of Nigeria, Nsukka (UNN). Samples of the leaves were later deposited at the University of Nigeria Herbarium (UNH) with herbarium code/number of 466. The collected plant leaves were dried at the laboratory under normal room temperature, which vary between 23°C and 28°C, for 20 days and thereafter grinded to powdered form.

Preparation of aqueous and ethanol extracts

Aqueous extract of *myrianthus arboreus* leaves (AEMA) was carried out by maceration of 1537.6 g of sample in 5 L of distilled water for 24 hours with 10 minutes continuous stirring at 6 hours interval. After that, the mixture was filtered, and the filtrate was concentrated using a water bath at 90°C, leaving the extract in a paste form. The percentage yield of the extract's concentration was 24.38%. Similarly, ethanol extract of *Myrianthus arboreus* (EEMA) was prepared according to previous method.[21] Here, 1275.3 g of sample was macerated in 4 L of 80% ethanol for 72 h, followed by filtration. A rotary evaporator at 40°C was used to concentrate the filtrate and thereafter evaporated on a water bath to dryness. The ethanol extract yield was 10.56%. Both extracts were weighed and stored until use.

Qualitative and quantitative Phytochemical analysis

Qualitative and quantitative phytochemical analyses were conducted on both extracts following already established standard procedures.[22-24]

Induction of diabetes mellitus

Induction of diabetes in rats was done by a single intraperitoneal injection of 150 mg/kg body weight of an alloxan monohydrate solution in a standard saline solution.[25] The alloxan solution was freshly prepared and rats were fasted overnight (12 h) before induction.[25] Blood samples were collected from the tail vein using a tail clip 72 hours after alloxan administration, and glucose levels were checked using a glucometer. Rats exhibiting ≥200 mg/dl of blood glucose level (BGL) were taken as diabetic,[26] and included in the study. The treatment started 72 hours after the alloxan injection, as the rats were confirmed to be diabetic. Treatment immediately commenced as this was taken as the first day of treatment and continued for the next 28 days.

Experimental design

Forty-five healthy male Wistar rats weighing 110 – 190 g were purchased from the animal house of the Department of Pharmacology and Toxicology, University of Nigeria, Nsukka. The rats were housed in plastic cages in the Department of Anatomy Animal House, University of Nigeria, under standard environmental conditions of 22.5°C, 12 hours light-dark cycle, with 50% to 60% relative humidity. The rats had free access to rat chows and drinking water *ad libitum* and were allowed to acclimatize to their new laboratory condition two weeks before the study. Ethical approval was gotten from the Research Ethics Committee of the Faculty of Basic Medical Sciences, University of Nigeria, Enugu campus, with approval number 076/08/2019. The experimental rats were maintained in line with the Ethics Committee's guidelines. The rats were randomly selected into nine groups each consisting of five rats (Table 1).

The alloxan-induced diabetic rats were treated with AEMA, EEMA, and Glibenclamide solutions orally by gastric gavage for 28 days. The rats' body weight (by digital electronic scale) and fasting blood glucose levels (BGL) using a commercial glucometer were measured weekly on the 7th, 14th, 21st, and 28th day of the study. Blood samples for the BGL were obtained from the tail veins of rats. Rats were fasted 12 hours after the last treatment before sacrifice by cervical decapitation under ether anesthesia.

Table 1: Experimental design

Group	Treatment
A: Normal control	1 mL of normal saline
B: Negative control	Alloxan and 1 mL of normal saline
C: Positive control	Alloxan and 5 mg/kg of glibenclamide (standard antidiabetic drug)
D: 100 mg/kg AEMA	Alloxan and 100 mg/kg of AEMA
E: 200 mg/kg AEMA	Alloxan and 200 mg/kg of AEMA
F: 400 mg/kg AEMA	Alloxan and 400 mg/kg of AEMA
G: 100 mg/kg EEMA	Alloxan and 100 mg/kg of EEMA
H: 200 mg/kg EEMA	Alloxan and 200 mg/kg of EEMA
I: 400 mg/kg EEMA	Alloxan and 400 mg/kg of EEMA

Excision of the pancreas and histological studies

The peritoneum of sacrificed animals was opened, and the pancreas was quickly harvested, weighed, and preserved in phosphate-buffered formalin (10%) for 48 hours. The pancreases were prepared for histological examination according to methods descried by Okoro,[27] and Salih et al.[28] Tissue sections of about 5 µm thickness were stained with hematoxylin and eosin (H and E) for the histological study. The prepared tissue slides were examined using a compound light microscope (Motic™) with ×4 ×10 and ×40 objective lenses. Motic™ 2.0. megapixel microscope camera (at ×100 and ×160 magnifications) was used to capture the photomicrographs.

Statistical analysis

Data were analyzed using EXCEL (Microsoft Office 16) and SPSS (IBM version 22) software. The results were statistically analyzed using one-way analysis of variance (ANOVA) at $p < 0.05$ with the post-hoc Tukey test. The data were presented as mean ± standard deviation.

Results and Discussion

Phytochemical constituents of Myrianthus arboreus leaf extracts Table 2 shows the qualitative phytochemical analysis of AEMA a**n**d EEMA. The qualitative study indicates the presence of saponins, glycosides, alkaloids, steroids, tannins, flavonoids, phenol, and terpenoids in both extracts. Figure 1 shows the concentrations of alkaloids, glycosides, and saponins present in the extracts with glycoside having the least value of 0.40 mg/g in both extracts. Some of these phytochemicals and antioxidants have also been confirmed in the aqueous extract of *Myrianthus arboreus* leaves.[29]Similarly, methanolic and aqueous extracts of *Myrianthus arboreus* stem bark have been reported with low glycosides, phenols, saponins, alkaloids, and flavonoids.[30]

Table 2: Qualitative phytochemical constituents of AEMA and EEMA

Phytochemicals	AEMA	EEMA

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

Saponins	+	+
Tannin	+	+
Flavonoid	+	+
Glycosides	+	+
Alkaloids	+	+
Steroids	+	+
Terpenoids	+	+
Phenol	+	+
Resin	–	–

– Absent; + Present in moderately high concentration

Effect of extract on body weight

The effect of both AEMA and EEMA on the rats' body weight and relative pancreas is presented in Table 3. The results show an increase in body weight of the standard control, positive control, and AEMA and EEMA treated groups in comparison with the untreated diabetic group. A significant decrease ($p < 0.05$) in body weight of the untreated group was well noticed with average value ranging from 171.7±16.3 g after induction to 150.7±32.2 g on the 28th day (last day) of the study. The increase in body weight of the standard control and positive control groups was significant ($p < 0.05$). Similarly, a significant ($p < 0.05$) increase in body weight was noted in the AEMA and EEMA treated groups. This increase in weight caused by AEMA and EEMA in treated rats was comparable to that of the positive control group treated with glibenclamide. No significant difference was observed between the AEMA groups at 200 and 400 mg/kg treatment and the standard control and positive control groups. For EEMA treatment, significant differences exist between the EEMA groups and those of the standard and positive control groups. Figure 2 compares the initial body weights with the final body weights. The pancreas weights of the diabetic groups show a significant decrease ($p < 0.05$) in comparison to that of the non-diabetic group. A significant decrease ($p < 0.05$) in pancreas weight was observed in the 400 mg/kg AEMA, 100 mg/kg EEMA, and 200 mg/kg EEMA treated groups compared to standard control, negative control, and positive control groups. AEMA at 200 mg/kg dose and EEMA at 400 mg/kg dose showed no significant ($p > 0.05$) restoration of pancreas weight when compared to the negative control group. The pancreas weights between these two groups did not differ significantly.

Table 3: Effects of extracts on body and pancreas weights

Group	Initial body weight (g)	Final body weight (g)	Pancreas weight (g)	Pancreas/body weight ratio
A: Normal control	152.3±2.2^{*b}	202.9±8.8*	4.1±0.4	0.020
B: Negative control	171.7±16.3^{*a}	150.7±32.2^{*a}	3.3±0.3	0.022
C: Positive control	173.1±20.4^{*a}	200.3±28.2^{b*}	3.2±0.4	0.016
D: 100 mg/kg AEMA	138.5±18.2^{*b}	177.1±32.2^{*c}	3.1±1.4	0.018
E: 200 mg/kg AEMA	169.9±2.3^{*a}	208.5±11.7^{*b}	3.5±0.2	0.017
F: 400 mg/kg AEMA	177.9±12.1^{*a}	203.6±12.0^{*b}	2.8±0.3	0.014
G: 100 mg/kg EEMA	143.4±15.9^{*b}	166.0±33.8^{*c}	2.9±1.2	0.017
H: 200 mg/kg EEMA	148.3±7.4^{*b}	154.4±9.9^{*b}	2.8±0.1	0.018
I: 400 mg/kg EEMA	142.3±2.8^{*b}	183.2±14.6^{*b}	3.4±0.6	0.019

Values are presented as mean ± Standard Error. Values in the same column having different letters are statistically significant ($P < 0.05$).Values in the same row having different letters of the alphabet are statistically significant ($P < 0.05$)

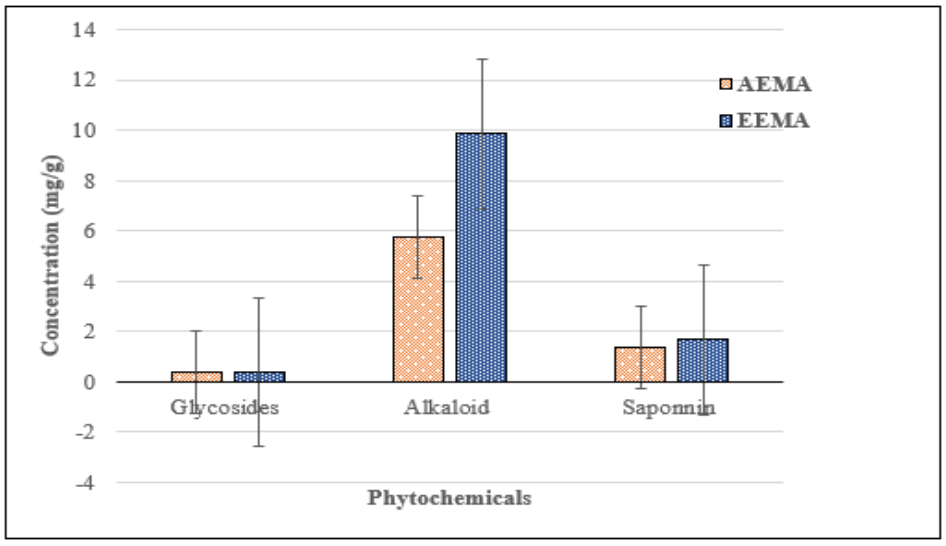

Figure 1: Concentration of glycosides, alkaloids and saponins in *Myrianthus arboreus* leaf extract

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

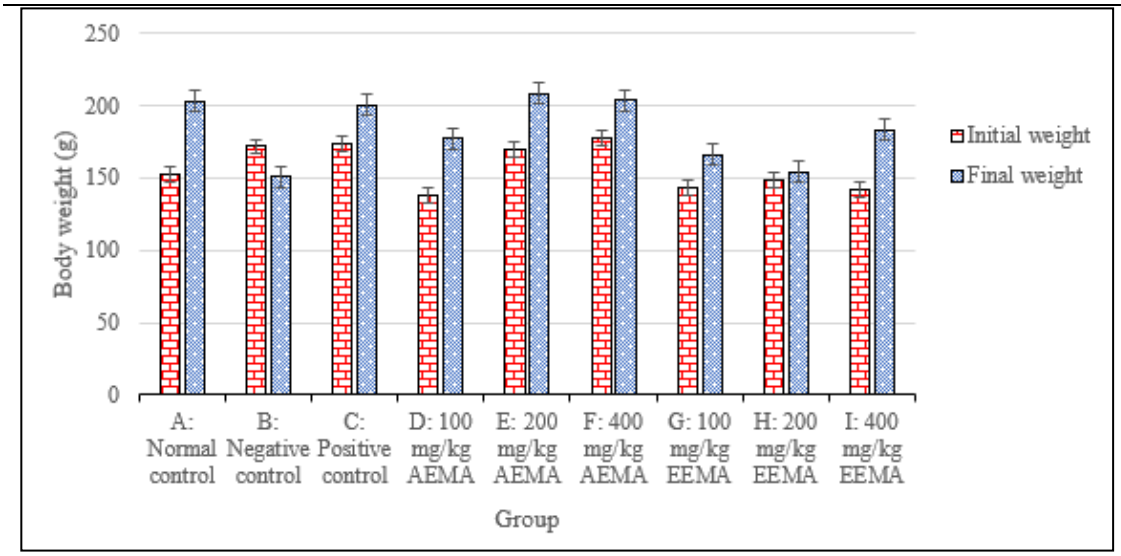

Figure 2: Comparison of initial and final body standard control, negative control, and treated groups

DM, a metabolic disorder, can impact the rate of nutrient metabolism and bioavailability thereby interfering with body growth and development,[31] hence, the noticeable weight loss in the untreated diabetic group. Several studies have reported the risks associated with a diabetic-related decrease in body weight and bone mass.[31,32]Like every other medicinal plant reported in the literature,[1,9,33] AEMA and EEMA show essential and nonessential nutritional elements that can boost body weight gain requirements of diabetic patients as evident in this study. Similar observations of weight gain in rats treated with an aqueous extract of *Myrianthus arboreus* leaves have also been reported.[26,29,34] The decrease in pancreatic weight of the diabetic groups noted in this study collaborate earlier reported studies of different medicinal plants.[35] Reduced pancreatic size is associated with type 1 and type 2 DM.[36,37] Several authors have linked such association to immunological abnormalities and diminished insulin production. [32,33] Though the relationship between pancreas size and weight and its insulin-secreting ability has not well been established, it is assumed that diminished pancreas weight and size may reduce insulin secretion, and patients with a small-sized pancreas are likely to develop diabetes.[38]

Effect of extracts on fasting blood glucose level

The fasting blood glucose levels (mg/dL) of the rats measured at different stages of the study are presented in Table 4. Figure 3 shows the comparison of the BGL (mg/dL) of treated groups with that of standard control and untreated groups. No significant change exists in the BGL of the standard control group throughout the period of the study. There was a significant change in the BGL of the diabetic groups.

Table 4: Effects of extracts on fasting blood glucose level

Group	Fasting blood glucose (mg/dL)					
	Pre-induction	**Induction**	**Week 1**	**Week 2**	**Week 3**	**Week 4**
A: Normal control	75.88±2.50	76.28±3.20	78.98±3.21	80.80±2.75	77.95±3.37	78.23±4.43
B: Negative control	73.03±3.88	262.55±32.42^{a}	286.15±17.81^{a}	300.85±19.27^{a}	333.30±50.30^{a}	360.25±58.41^{a}
C: Positive control	80.18±2.89	269.70±70.30^{*b}	224.80±36.94^{*b}	175.18±22.48^{*b}	107.33±8.49^{*b}	79.00±3.94^{*b}
D: 100 mg/kg AEMA	74.15±4.57	271.75±24.38^{*c}	225.25±23.69^{*c}	178.73±7.36^{*c}	107.78±13.20^{*c}	88.93±8.98^{*c}
E: 200 mg/kg AEMA	86.13±6.11	284.05±52.48^{*c}	229.95±44.14^{*c}	176.43±28.60^{*c}	102.05±10.14^{*c}	85.13±6.85^{*c}
F: 400 mg/kg AEMA	76.18±3.54	264.30±15.34^{*c}	222.38±24.76^{*c}	152.48±35.30^{*c}	114.60±21.54^{*c}	94.78±14.52^{*c}
G: 100 mg/kg EEMA	79.03±6.11	271.48±62.24^{*d}	242.65±58.32^{*d}	178.78±39.20^{*d}	133.23±18.93^{*d}	89.63±6.05^{*d}
H: 200 mg/kg EEMA	80.08±10.25	260.50±35.29^{*d}	214.80±13.81^{*d}	183.60±25.75^{*d}	100.63±6.21^{*d}	86.05±11.35^{*d}
I: 400 mg/kg EEMA	74.63±6.76	248.73±46.44^{*d}	223.40±21.82^{*d}	190.08±15.84^{*d}	113.23±15.43^{*d}	86.00±9.23^{*d}

Values with different superscript letters in the same column are significantly different at $P < 0.05$. * ($P < 0.05$) comparison with normal control; ^{b} ($P < 0.05$) positive control with negative control group; ^{c} ($P < 0.05$) AEMA groups with negative control group; ^{d} ($P < 0.05$) EEMA groups with negative control group

In the untreated group, induction of diabetes caused a significant increase ($p < 0.05$) in BGL from 73.03 ± 3.88 mg/dL (pre-induction) to 262.55±32.42 mg/dL (induction) and progressively to 360.25 ± 58.41 mg/dL (week 4). Treatment with glibenclamide, AEMA, and EEMA shows a progressive significant ($p < 0.05$) depression of the BGL across the study period. The positive control group (treatment with glibenclamide) shows a significant decrease in BGL from 269.70±70.30 mg/dl (induction) to 79.00 ± 3.94 mg/dL (week 4) compared to 80.18 ± 2.89 mg/dL of pre-induction. AEMA and EEMA also indicated a similar decrease. Treatment with AEMA and EEMA was statistically significant ($p < 0.001$) among the groups as well as in comparison with the positive control group but with no dependence on doses. However, the most probable reduction was observed in groups treated with 200 mg/kg AEMA, and 100 and 200 mg/kg EEMA, which caused about 70.0% and 67.0% reduction respectively. The ability of AEMA and EEMA to lower BGL shows that the treated rats had better chances of glucose utilization. This further suggests that AEMA and EEMA may exert an insulin-like effect on peripheral tissues by

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

promoting glucose uptake or glucose absorption into muscles, hindering hepatic gluconeogenesis and adipose tissues.[39]

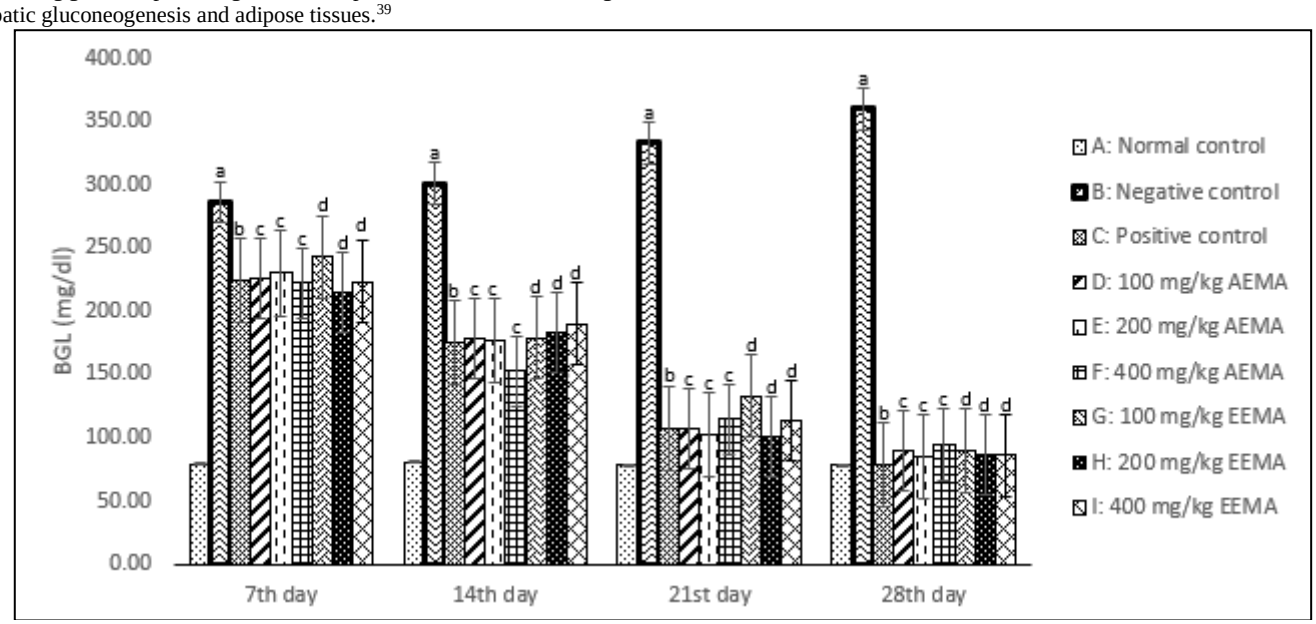

Figure 3: Comparison of blood glucose level (mg/dL) of treated groups with standard control and untreated groups for 28 days (4 weeks). [a]P< 0.05 (regular control group with untreated diabetic group), [b]P< 0.05 (glibenclamide treated group with untreated diabetic group) [c]P< 0.05 (AEMA treated group with untreated diabetic group) and[d]P< 0.05 (EEMA treated group with untreated diabetic group)

The observed reduction in BGL of the diabetic groups treated with AEMA and EEMA confirmed previously reported anti-hyperglyceamic effect of *Myrianthus arboreus*. Several plants have effectively lowered blood glucose level and have long been applied as alternative substitution in diabetes management.[40] Like other plant extracts, *Myrianthus arboreus* leaves extracts of this present study have demonstrated potency in ameliorating BGL in alloxan-induced diabetic rats. Other plant parts have also demonstrated this potency as reported in other studies.[20,40] This antidiabetic property can be credited to the existence of flavonoids and other phytochemicals. It is revealed that flavonoids have positive health effects on metabolic disorders, such as diabetes.[41] AEMA and EEMA of present study exhibited the antidiabetic and anti-hypoglycaemic potentials by enhancing insulin secretion and glucose homeostasis modulate. These abilities of glucose homeostasis modulation and exertion of hypoglyceamic and anti-hyperlipidaemic activities on diabetic animals have been noted using the root bark extracts of this same plant.[20,39] The hypoglyceamic effect demonstrated by AEMA and EEMA can be credited to glycosides, which help to adjust glucose transported 4 (GLUT4) expression and tyrosine phosphorylation of insulin receptors.[40,42] Also, antioxidants such as flavonoids are known in reducing oxidative stress, hence the noticeable progressive decrease in diabetic complications of the treated rats. In a similar study,[43] stem bark extracts of *Myrianthus arboreus* had been reported to reduce BGL levels of experimental animals to near-average values. Based on this current study and others earlier reported, *Myrianthus arboreus* can be designated as a potent antidiabetic plant.

Effect of extracts on pancreatic histology

The histological examinations of the pancreatic sections stained with hematoxylin and eosin (H & E) of the standard control (plate A), negative control (plate B), and positive control (plate C) are shown in Figure 4. Figures 5 and 6 show the histological examinations of the pancreatic sections (H & E) of the AEMA at 100, 200, and 400 mg/kg (plates D, E, and F) and EEMA at 100, 200, and 400 mg/kg (plates G, H and I), respectively. The pancreatic section of the standard control group shows a typical pancreas structure with whole islets consisting of centrally positioned beta cells. The control group shows a standard interstitial architecture, very compact in appearance, and encircled by seroacinar cells. These features show intact pancreatic islets cells. In the other groups, induction of diabetes through alloxan administration caused severe histopathological changes in the examined pancreas. Reduction in number and hypocellularity of islets of Langerhans were prominent in the diabetic groups. In the negative control group (Figure 4, Plate B), noticeable distortion of islet and acini cells was observed. Features such as necrotic changes of pancreatic islets, size reduction, karyolysis, nuclear disappearance, and visible residue of wrecked beta cells were observed. Administration of glibenclamide, AEMA, and EEMA showed regeneration of pancreatic islets and restoration of beta cells. The acini cells looked basophilic. Plates D, E, and F (Figure 5) for AEMA at 100, 200, and 400 mg/kg, respectively, and plates G, H, and I (Figure 6) for EEMA at 100, 200, and 400 mg/kg respectively showed evidence of improved restoration of beta cells with more islet regeneration in comparison with the untreated diabetic group. Minimal necrosis was noted in groups treated with AEMA and EEMA. Treatment with AEMA shows a gradual increase in pancreas islet size compared to EEMA. Diabetes is associated with progressive disruption to destruction of islet cells that produce insulin.[44,45] The decrease in pancreatic beta cells indicates gradual impairment of insulin secretion,[11,23] resulting in a functional decrease of the beta cells and insulin resistance. This is evident in the untreated group. The histopathological study revealed the ability of AEMA and EEMA in restoring lost islet cells and beta cell function of pancreas of treated rats. When compared with the untreated case, the gradual increase of the pancreas islet area of AEMA and EEMA treated groups indicates the regeneration of new islets beta cells. Alloxan induces diabetes by weakening and progressively destroying pancreas insulin-secreting cells (beta cells) with the consequential effect of hypoinsulinemia and hyperglyceamia.[44] Reduction and destruction of the beta cells can result in deficiency of insulin needed for proper glucose metabolism thereby leading to hyperglyceamia. The restoration and increase in beta cells initiated by AEMA and EEMA show upgraded insulin secretion activity in the treated group. The gradual increase in beta cells and pancreas islet area of this study corroborates earlier findings of reduced blood glucose levels. This ability of AEMA and EEMA could be attributed to the phytochemical contents, such as flavonoids and phenols with proven antioxidant properties. Though the mechanism of action is not well known, it has been established that the flavonoid fraction of *Myrianthus arboreus* increases beta cell population and reduces blood glucose.[43] Also, the phenolic content of medicinal plants positively contributes to the antioxidant activities exhibited by their extracts. The phenolic constituent of this plant may have stopped further destruction of the remaining beta cells by cleaning up the circulating reactive oxygen species generated by the alloxan, thus allowing other phytochemicals to induce regenerative activities.[42,43]

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

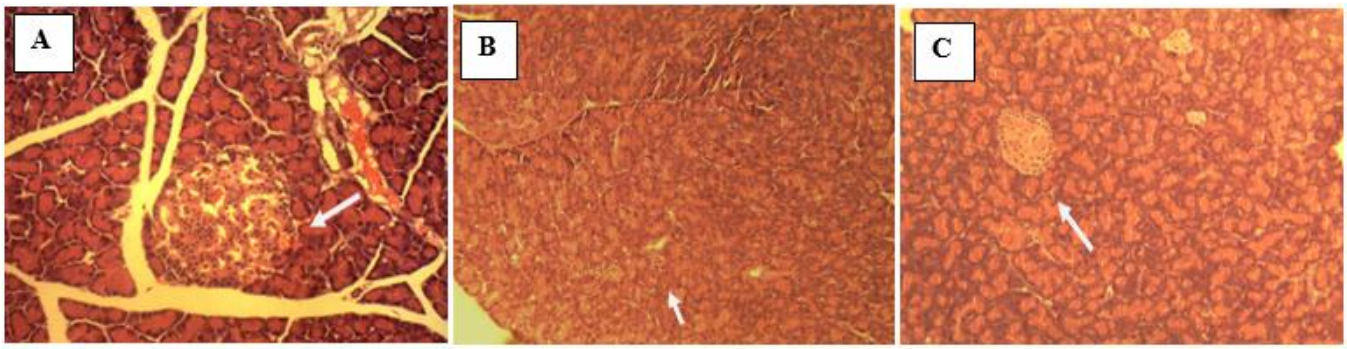

Figure 4: Histopathological examination of the pancreas (H & E): (A) Normal control groups with regular islet section. Pancreas cells were present in their average proportion; (B) Diabetic negative control group having reduced islet section and interstitial congestion; (C) positive control with normal interstitium and islet section

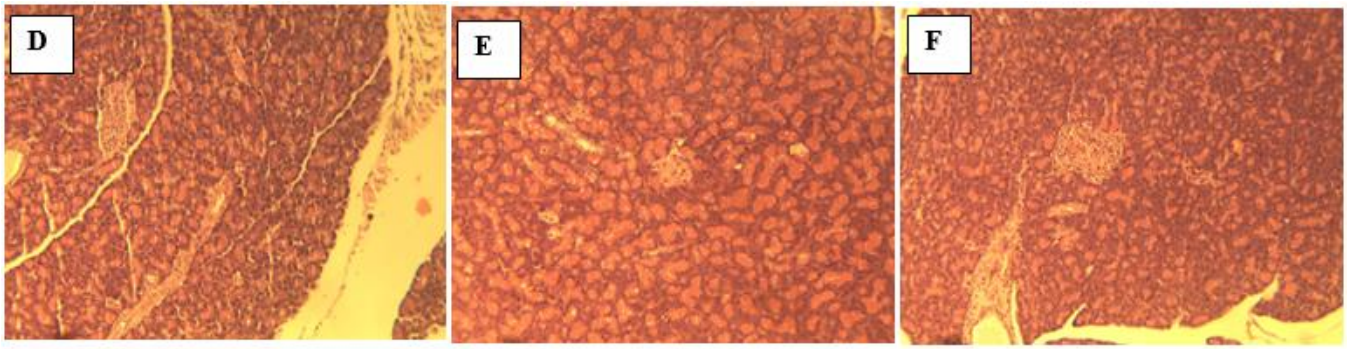

Figure 5: Histopathological examination of the pancreas (H & E) showing slides from (D, E, and F) AEMA treated group (at 100, 200, and 400 mg/kg respectively) showing regeneration of pancreas islet and beta cells

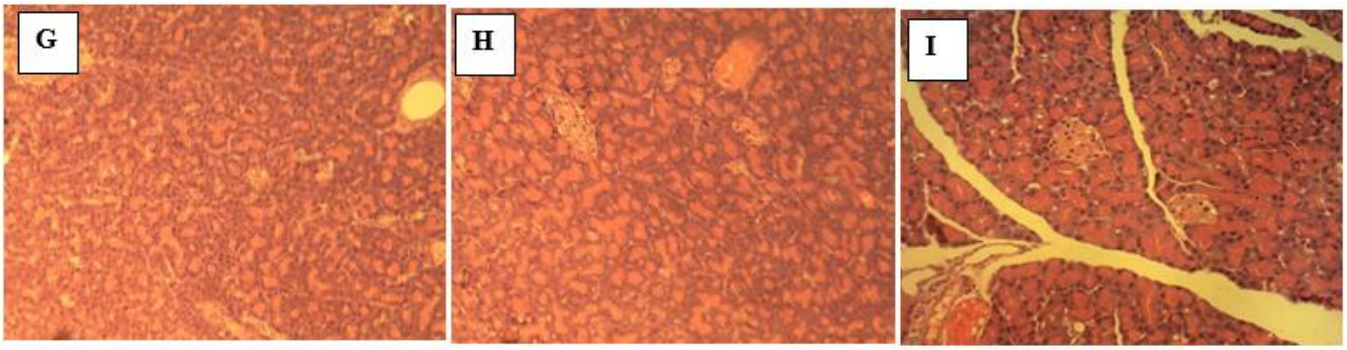

Figure 6: Histopathological examination of the pancreas (H & E) showing slides (G, H, and I) EEMA treated group (at 100, 200, and 400 mg/kg respectively) showing regeneration of pancreas islet and beta cells

Conclusion

Myrianthus arboreus possesses antidiabetic properties, which may be connected to presence of phytochemicals and its antioxidant activity. The results of the study support using aqueous and ethanol extracts of the *Myrianthus arboreus* leaves as an herbal remedy for treating diabetes. The study shows positive prospect of the plant in alleviating high blood glucose level and restoring pancreas beta cells. Its pharmaceutical complexity in relation to physiological factors is recommended for future study.

Conflict of interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

References

1. Assadi S, Shafiee SM, Erfani M, Akmali M. Antioxidative and antidiabetic effects of *Capparis spinosa* fruit extract on high-fat diet and low-dose streptozotocin-induced type 2 diabetic rats. *Biomed Pharmacother*. 2021; 138:111391. https://doi.org/10.1016/j.biopha.2021.111391

2. Iwara IA, Mboso EO, Ibor OR, Elot K, Igajah C, Bassey AA, Eteng OE, Mgbeje BIA, Igile GO, Eteng MU, Arukwe A. Modulatory effects of extract of Heinsia crinita against fructose/ streptozotocin-induced oxidative stress in diabetic

rat models *Heliyon.* 2023; 9:e21308.https://doi.org/10.1016/j.heliyon.2023.e21308

3. Hashemnia M, Oryan A, Hamidi AR, Mohammadalipour A. Blood glucose levels and pathology of organs in alloxan-induced diabetic rats treated with hydroethanol extracts of *Allium sativum* and *Capparis spinosa. Afr J. Pharm Pharm.* 2012; 6(21):1559–1564.https://doi.org/10.5897/AJPP12.330
4. WHO World Health Organization: Global Status Report on noncommunicable diseases. 2014. Accessed from: https://iris.who.int/bitstream/handle/10665/148114/9789241564854_eng.pdf?sequence=1
5. Abegunde DO, Mathers CD, Adam T, Ortegon M, Strong K. The Burden and Costs of Chronic Diseases in Low-income and Middle-income Countries. *The Lancet*. 2007; 370(9603):1929-1938. https://doi.org/10.1016/S0140-6736(07)61696-1
6. Raheleh A, Mokarram P, Dastghaib S, Darbandi S, Darbandi M, Zal F, Akmali M, Ranjbar-Omrani GH. Hypoglycemic effect of aquatic extract of Stevia in pancreas of diabetic rats: PPARγ-dependent regulation or antioxidant potential. *Avicenna J. Med Biotechnol*. 2016; 8(2):65–72.
7. Patel V and Sharma V. Antioxidant and antidiabetic effect of *Capparis spinose* linn.instreptozotocin induced diabetic rats. EJBPS. 2015; 2:308–318.
8. Obasi DC, Ogugua VN. GC-MS analysis, pH and antioxidant effect of Ruzu herbal bitters on alloxan-induced diabetic rats. *BiochemBiophy* Rep. 2021; 27:101057.https://doi.org/10.1016/j.bbrep.2021.101057
9. Mohammed A, Tajuddeen N. Antidiabetic compounds from medicinal plants traditionally used for treating diabetes in Africa: A review update (2015-2020). *South Afr J. Bot.* 2022; 146:585-602. https://doi.org/10.1016/j.sajb.2021.11.018
10. Chikezie PC, Ojiako OA, Nwufo KC. Overview of Antidiabetic Medicinal Plants: The Nigerian Research Experience. *J Diabetes Metab*. 2015; 6:546.
11. Abbas G, Al-Harrasi A, Hussain H, Hamaed A, Supuran CT. The management of diabetes mellitus-imperative role of natural products against dipeptidyl peptidase-4, a-glucosidase and sodium-dependent glucose cotransporter 2 (SGLT2). *Bioorg Chem.* 2019; 86:305–315. https://doi.org/10.1016/j.bioorg.2019.02.009
12. Edem DO, Edagha IA, Ette BB, Agwuigwo PE. Effects of *Tapinanthus globiferus* leaf extract on blood glucose and pancreatic histology in alloxanized and normoglycemic rats. Arc *Diabete Endocr Sys.* 2020; 3(2):34–43.
13. Olonode ET, Aderibigbe AO, Bakre AG. Antinociceptive activity of the crude extract of *Myrianthus arboreus* P.Beauv (Cecropiaceae) in mice. *J Ethnopharmacol*. 2015; 171:94–98.http://dx.doi.org/10.1016/j.jep.2015.05.005
14. Burkill HM. The useful plants of West Tropical Africa. Families J L. 1985; 3. Royal Botanic Garden, Kew.
15. Ojinnaka CM, Okogun JI, Okorie DA. Triterpene acids from *Myrianthus arboreus*. *Phytochem*. 1980; 19(11):2482-2483.
16. Ngounou FN, Lontsi D, Sondengam BL. A pentacyclic triterpene diacid from *Myrianthus arboreus. Phytochem.* 1988; 27(7):2287-2289.
17. Otitoju G, Nwamarah J, Otitoju O, Odoh E, Iyeghe L. Phytochemical composition of some underutilized green leafy vegetables in Nsukka urban LGA of Enugu State. J Biodiv Env Sci. 2014; 4:208-217.
18. Seukep JA, Ngadjui B, Kuete V. Antibacterial activities of *Fagaramacrophylla, Canariumschweinfurthii, Myrianthus arboreus, Dischistocalyx grandifolius and Tragia benthamii* against multi-drug resistant Gram-negative bacteria. *Springerplus*. 2015; 4(1):567.
19. Kasangana P, Haddad P, Stevanovic T. Study of polyphenol content and antioxidant capacity of *Myrianthus arboreus* (*Cecropiaceae*) root bark extracts. Antioxidants. 2015; 4(2):410-426.
20. Kasangana PB, Nachar A, Eid HM, Stevanovic T, Haddad PS. Root bark extracts of *Myrianthus arboreus P. Beauv*. (*Cecropiaceae*) exhibit antidiabetic potential by modulating hepatocyte glucose homeostasis J. Ethnopharm. 2018; 211:117–125 http://dx.doi.org/10.1016/j.jep.2017.09.017
21. Boye A, Barku VYA, Acheampong DO, Ofori EG. *Abrus precatorius* leaf extract reverses alloxan/nicotinamide-induced diabetes mellitus in rats through hormonal (insulin, GLP-1, and glucagon) and enzymatic (α-amylase/α-glucosidase) modulation BioMed Res Int. 2021; 2021:9920826. https://doi.org/10.1155/2021/9920826
22. Harborne, J. B. Phytochemical Methods: A Guide to Modern Techniques in Plant Analysis New York: Chapman and Hall. 1993 p. 22.
23. Boham BA, Kocipai AC. Flavonoids and condensed tannins from the leaves of Hawaiian *Vaccinum vaticulatum* and *V. calycinum. Pacific Sci*. 1994; 48:458–463.
24. Erukainure OL, Ebuehi OAT, Adeboyejo FO, Okafor EN, Muhammad A, Elemo GN. Fiber-enriched biscuit enhances insulin secretion, modulates b-cell function, improves insulin sensitivity, and attenuates hyperlipidemia in diabetic rats. *PharmaNutr*. 2013; 1:58–64.http://dx.doi.org/10.1016/j.phanu.2013.02.001
25. Owa SO, Taiwo AA, Okosun JA, Otohinoyi DA, Akujobi YO, Oyewale DG, Ibraheem O, Edewor-Ikupoyi TI, Adeyemi OS. The biochemical effects of lime concentrate 'Aporo' and *Mucunapruriens* seeds extract on Alloxan-induced diabetic rats. *J. Taibah Uni Med Sci.* 2016; 11(3):260-267. http://dx.doi.org/10.1016/j.jtumed.2016.02.004
26. Obiandu C and Green K. Assessment of the Anti-Diabetic and Some Biochemical Effects of *Myrianthus aboreus* on Wistar Rats. Saudi J. Med. 2019; 4(2):142-146. DOI: 10.36348/sjm.2019.v04i02.013
27. Okoro I. Histological techniques. In: Manual of practical histology. 2002; pg 4-9 2nd Ed. Peace Publishers Ltd, Owerri.
28. Salih ND, Azmi N, Gopalan HK. The protective effects of *Phaleria macrocarpa* leave methanol extract on pancreatic islets histology in streptozotocin-induced diabetic rats. SciInt (Lahore). 2015; 27(5):4219-4224.
29. Onyechi AU, Ibeanu VN, Maduforo AN, Ugwuonah AU, Nsofor LD. Effects of *Corchorus olitorius, Myrianthus arboreus* and *Annona muricata* aqueous leaves extracts on body weight, blood glucose levels and lipid profile of alloxan-induced diabetic rats. J. Diet Ass Nig. 2018; 9:11–20.
30. Biapa PCN, Agbor GA, Oben JE, Ngogang JY. Phytochemical studies and antioxidant properties of four medicinal plants used in Cameroon. Afr J. TradCompl Alt Med. 2007; 4(4):495 – 500.
31. Erukainure OL, Ebuehi OAT, Adeboyejo FO, Aliyu M, Elemo GN. Hematological and biochemical changes in diabetic rats fed with fiber-enriched cake. J Acute Med. 2013; 3:39-44. http://dx.doi.org/10.1016/j.jacme.2013.03.001
32. Fasola TR, Ukwenya B, Oyagbemi AA, Omobowale TO, Ajibade TO. Antidiabetic and antioxidant effects of *Croton lobatus L.* in alloxan-induced diabetic rats. *J Intercult Ethnopharmacol*. 2016; 5(4):364–371.
33. Aleem A, Shahnaz S, Javaid S, Ashraf W, Rasool MF, Ahmad T, Alotaibi AF, Albeshri KS, Alqahtani F, Imran I. Chronically administered *Agave americana var. marginata* extract ameliorates diabetes mellitus, associated behavioral comorbidities and biochemical parameters in alloxan-induced diabetic rats. *Saudi Pharm J*. 2022; 30:1373–1386. https://doi.org/10.1016/j.jsps.2022.06.003
34. Awounfack CF, Ateba SB, Zingue S, Mouchili OR, Njamen D. Safety evaluation (acute and sub-acute studies) of the aqueous extract of the leaves of *Myrianthus arboreus P.*

Beauv. (*Cecropiaceae*) in Wistar rats. J Ethnopharm. 2016; 194:169–178. http://dx.doi.org/10.1016/j.jep.2016.08.052

35. Arokoyo DS, Oyeyipo IP, Du Plessis SS, Chegou NN, Aboua YG. Modulation of inflammatory cytokines and islet morphology as therapeutic mechanisms of Basella alba in
36. Erfani MN, Tabandeh MR, Shahriari A, Soleimani Z. Okra (*Abelmoscus esculentus*) improved islets structure, and down-regulated PPARs gene expression in pancreas of high-fat diet and streptozotocin-induced diabetic rats. Cell J. 2018; 20(1):31–40. doi:10.22074/cellj.2018.4819.
37. Yagihashi S. Diabetes and pancreas size, does it matter? J *Diabetes Investig.* 2017; 8(4):413–415.
38. Dickson RA, Harley BK, Berkoh D, Ngala RA, Titiloye NA, Fleischer TC. Antidiabetic and haematological effect of *Myrianthus arboreus* P. Beauv. Stem bark extract in streptozotocin-induced diabetic rats. *Int J. Pharm Sci Rev* Res. 2016; 7:4812–4826.
39. Maithani A, Parcha V, Pant G, Kumar D, Dhulia I. Evaluation of anti-hyperglycaemic action of different fractions and sub-fractions from aqueous extract of aloe vera Linn. leaf on alloxan Induced type 2 diabetic rats. *Asian Pac J Trop Biomed*. 2012; S1691-S1695.
40. Middleton E, Kandaswami C, Theoharides TC. The effects of plant flavonoids on mammalian cells: Implications for inflammation, heart disease, and cancer. *Pharm Rev*. 2000; 52:673–751. PMID:11121513
41. Oladele SB, Ayo JB, Adaudi AO. Medicinal and physiological properties of flavonoids, coumarine derivatives and antheraquinones of plant origin. *W Afr J. Pharmacol Drug Res*. 1995; 11:134-144.
42. Harley, BK, Dickson RA, Amponsah IK, Ben IO, Adongo DW, Fleischer TC, Habtemariam S. Flavanols and triterpenoids from *Myrianthu sarboreus* ameliorate hyperglycaemia in streptozotocin-induced diabetic rats possibly via glucose uptake enhancement and a-amylase inhibition. *Biomed Pharmacother*. 2020; 132:110847.
43. Tang SC, Shen CN Lin PY, Peng SJ, Chien HJ, Chou YH, Chamberlain CE, Pasricha PJ. Pancreatic neuro-insular network in young mice revealed by 3D panoramic histology. *Diabetologia.* 2018; 61:158–167. DOI 10.1007/s00125-017-4408-y
44. Ujowundu CO, Onyema CR, Nwachukwu N, Ujowundu FN, Onwuliri VA, Igwe KO, Achilike JJ, Udensi JU. Antioxidative effect of phenolic extract of *vitex doniana* leaves on alloxan-induced diabetic stress and histological changes in the pancreas of Wistar rat. *Trop J Nat Prod Res.* 2022; 6(2):270-275. doi.org/10.26538/tjnpr/v6i2.1