Ethanol root extract and fractions of *Sphenocentrum jollyanum* abrogate hyperglycemia and low body weight in Streptozotocin-induced diabetic Wistar albino Rats.

[1]Ugwu Okechukwu Paul-Chima, [1,2*]Alum Esther Ugo, [1]Okon Michael Ben, [2,3]Aja Patrick M., [1]Obeagu Emmanuel Ifeanyi and [4]Onyeneke E.C.

[1]Department of Publication and Extension, Kampala International University Uganda.

[2]Department of Biochemistry Ebonyi State University Abakaliki, Nigeria.

[3]Department of Biochemistry Kampala International University Uganda.

[4]Department of Biochemistry University of Benin, Edo State Nigeria.

Corresponding author: Alum Esther Ugo*

Mailing Department of Publication and Extension, Kampala International University, Uganda.

Corresponding Author's e-mail esther.alum@kiu.ac.ug; esther.alum@ebsu.edu.ng

Authorship/contributions: The authors confirm contribution to the paper as follows:

Study conception and Design: Ugwu Okechukwu Paul-Chima and Onyeneke E.C; **Data collection**: Okon Michael Ben and Alum Esther Ugo; **Analysis and Interpretation of results**: Aja Patrick M; **Draft manuscript preparation**: Ugwu Okechukwu Paul-Chima and Onyeneke E.C; **Editing and revision of manuscript**: Alum Esther Ugo and Obeagu Emmanuel I.

All authors reviewed the results and approved the final version of the manuscript**.**

Funding: This research did not receive any specific grant from funding agencies in the public, commercial, or not for- profit sectors.

Conflict of interest

Authors declare no conflicts of interest in this publication.

Data availability

There is no data to share

ABSTRACT

Objectives: Effects of ethanol root extract and fractions of *Sphenocentrum jollyanum* on fasting blood glucose and body weight of streptozotocin-induced diabetic albino rats were carried.

Methods:48 rats were randomly assigned into 8 groups. Groups 1 and 2 were diabetic rats treated with 0.5 ml of normal saline and 0.5mg/kg b.w of glibenclamide, respectively while group 3 were non diabetic rats treated with 0.5ml of normal saline. Groups 4, 5 and 6 rats were diabetic rats treated with 250, 500 and 1000 mg/kg b.w of extract, respectively, while rats in groups 7 and 8 were diabetic rats treated with 250 mg/kg b.w of methanol and ethylacetate fractions of *Sphenocentrum jollyanum*, respectively. Diabetes was induced by intraperitoneal injection of a single dose of 70mg/kg b.w of streptozotocin. Fasting blood glucose levels were determined with glucometer.

Key findings: Treatment of diabetic rats with the extract and fractions of *Sphenocentrum jollyanum* at varied doses significantly ($P<0.05$) decreased glucose level in a dose and time dependent manner. There was an increase in body weights of rats during treatment.

Conclusion:The extract and fractions lowered glucose levels and raised body weights of diabetic rats. This suggests that *Sphenocentrum jollyanum* possess antidiabetic property and could be useful in the management of diabetes.

Keywords: *Sphenocentrum jollyanum*, Fasting blood glucose, albino rats, diabetes, streptozotocin

INTRODUCTION

Diabetes mellitus (DM) is a metabolic disorder resulting from a defect in insulin action, insulin secretion or both [1,2,3]. Insulin deficiency leads to chronic hyperglycemia with disturbances in fat, carbohydrate and protein metabolism. It is a global disease, prevailing throughout the world, although the prevalence rate differs from country to country [2]. The increasing ageing population, consumption of calorie-rich diet, obesity and sedentary lifestyle has led to tremendous increase in the number of hyperglycemics worldwide [4].

Herbal medicines are great body balancers and tonics that help in regulating some body functions. It can be used to support metabolic processes of the body and offer some other nutrients that the body fails to receive due to poor diet or environmental deficiencies in the soil. Interests in medicinal plants have been fueled by the rising costs of orthodox drugs in the maintenance of health and by the bio-prospecting of new plant derived drugs. Also, in spite of the presence of anti-diabetic drugs, remedies from medicinal plants are considered to be free from the side effects compared with synthetic ones [5]

Sphenocentrum jollyanum is an erect shrub that belongs to the family Menispermacea [6]. It is called "Ezeogwu" in Igbo, "Aduro Koroo" or "Okramankote" in the Akan Language in Ghana [7]. *Sphenocentrum jollyanum* has been shown to have antihypertensive, antioxidant, antinociceptive, antiviral, antianemic and anti-angiogenic effects in animals [7,8, 9]. The plant is also documented for its use against chronic coughs, worms and other inflammatory conditions as well as tumors [8,10]. The plant is traditionally used as remedy for feverish conditions as well and as an aphrodisiac [11,12,13]. Studies have shown that the leaves possess significant antipyretic and analgesic activities [13]. The roots and leaves have been reported to be active against polio [13]. Samuel *et al.* [14] and Sinbad *et al.* [15] reported the anti-inflammatory potentials of aqueous and ethanol leaf, root and stem-extracts of *Sphenocentrum jollyanum.*

Streptozotocin (STZ) is widely used to induce diabetes in various laboratory animals as it is particularly toxic to the pancreatic insulin-producing beta cells in mammals [12,16]. Streptozotocin inhibits insulin secretion and causes a state of insulin-dependent diabetes mellitus [16,17,18].

Despite the well-documented reports on the pharmacological activities of *Sphenocentrum jollyanum,* there is paucity of information about its anti-hyperglycemic effect using methanol and ethylacetate solvents. This informed the choice of methanol and ethylacetate as fractionating solvents in this study. Therefore, we aimed to ascertain the effects of ethanol root extract; methanol and ethylacetate fractions of *Sphenocentrum jollyanum* on fasting blood glucose and body weight of streptozotocin-induced diabetic albino rats.

MATERIALS AND METHODS

Collection of Biological Materials

The present study was carried out using the roots of *Sphenocentrum jollyanum* and albino rats and mice. Fresh roots of *Sphenocentrum jollyanum* were collected from Ovoko in Igbo-Eze South Local Government Area of Enugu State, Nigeria and was authenticated in the *Herbarium* Unit of Department of Botany, University of Nigeria, Nsukka by Mr O. Onyeukwu. Part of the authenticated plant was deposited in the *herbarium* for reference purposes (UNN/03620).

Eighty eight albino *wistar* rats and thirty two male albino mice were purchased from the Department of Zoology, University of Nigeria, Nsukka, Enugu State, Nigeria. They were acclimatized

for a period of two weeks at the animal house of the Department of Biochemistry, Ebonyi State University, Abakaliki, Nigeria prior to commencement of experiment. They were maintained at room temperature, 12hr day/night period and fed *ad libitum* on water and growers mash; weighed prior to commencement of experiment and daily till the end of the experiment.

Ethical Considerations

The study was approved on 15th February, 2018 by the Biochemistry Department Ethical Committee of Ebonyi State University Abakaliki, Nigeria (Ethical Approval number: EBSU/BCH/ET/18/003). The guidelines agree with world standard for care and use of laboratory animals in research (NIH Publication No. 85- 23, revised 1996).

Preparation of the Plant Extract: The roots of *Sphenocentrum jollyanum* were harvested and washed under tap water to remove contaminants and air dried under shade. They were pulverized using laboratory milling machine and sifted using 0.25 mm sieve. One thousand five hundred gram (1,500g) of the powdered root sample of *Sphenocentrum jollyanum* was soaked in 7500 ml of ethanol for 48 hours with agitation. The resulting ethanol root extract was filtered using muslin cloth and evaporated to dryness using rotary evaporator at a temperature of 45^0C. The concentrated ethanol root extract of *Sphenocentrum jollyanum* was used for subsequent analyses.

Fractionation of the Crude Extract of *Sphenocentrum jollyanum* Roots: The ethanol root extract of *Sphenocentrum jollyanum* (20 g) was fractionated in a glass column (150 cm x 1.5 cm) packed with 200 g of a slurry of silica gel G. (70-230 mesh). The column was eluted in succession with 500 ml ethyl acetate and 500 ml methanol to obtain ethyl acetate (EAF) and methanol (MF) fractions respectively. The resulting fractions were evaporated to dryness using rotary evaporator at a temperature of 45^0C. The concentrated ethyl acetate (EAF) and methanol root fractions of *Sphenocentrum jollyanum* were used for subsequent analyses.

Determination of acute Toxicity Studies (LD_{50}): Acute toxicity studies (LD_{50}) was determined using [19] method.

Induction of Diabetes: The baseline blood glucose levels were determined before the induction of diabetes. Rats were fasted overnight and experimental diabetes induced by intraperitoneal injection of streptozotocin (STZ) with a single dose of 70mg/kg body weight. STZ was dissolved in 0.1M citrate buffer at pH of 4.5 [18]. After three days, rats with blood glucose level greater than 250mg/dl that exhibit hyperglycemia were selected for the experiment [16]. The Accu-Check one-touch blood glucose monitoring meter and test strips were used for the assay.

Fasting Blood Glucose: The fasting blood glucose levels were determined with glucometer (Accu-Check one-touch blood glucose monitoring meter and test strips) after fasting the rats for 12h.

Statistical analysis: Results were expressed as mean± standard deviations where applicable. The data were subjected to one-way analysis of variance (ANOVA), followed by Post hoc Duncan multiple comparison test using SPSS software version 21 and $p < 0.05$ was regarded as significant.

RESULTS

The LD_{50} of the crude ethanol root extract of *Sphenocentrum jollyanum* in this study is greater than 5000 mg/kg body weight (Table 1) meaning that the crude ethanol root extract is not toxic at this dose.

<Insert Table 1>

Effect of Crude Ethanol Root-Extract and Fractions of *Sphenocentrum jollyanum* on Glucose Level in STZ-induced Diabetic Albino Rats

The treatment of STZ-induced diabetic albino rats with ethylacetate and methanol fractions of *Sphenocentrum jollyanum* at doses of 100, 150, 200, 250 and 300 mg/kg body weight significantly ($p<0.05$) decreased the blood glucose level in dose and time dependent manner as shown in figures 1 and 2. Treatment with crude ethanol root-extract at doses of 250, 500 and 1000 mg/kg body weights and 250 mg/kg body weight of ethylacetate and methanol root fractions significantly ($p<0.05$) decreased the blood glucose level in dose and time dependent manner as shown in figure 3. The result equally showed that on day 21 of treatment, the blood glucose level of rats treated with 500 and 1000 mg/kg body weights of ethanol crude root extract and 250mg/kg body weight of ethylacetate and methanol fractions decreased significantly ($p<0.05$) when compared with the positive control rats. But the effect on the standard control was quite similar to that of the crude extract and fractions on day 21 of treatment. The result also showed that there was no significant ($p>0.05$) changes in the glucose level across the groups after the twenty one days of treatment with both 1000 mg/kg body weight of the extract and 250 mg/kg body weight of the fractions as shown in figure 3.

<Insert Figure 1, 2 and 3>

Effect of Crude Ethanol Root-Extract and Fractions of *Sphenocentrum jollyanum* on Body Weight in STZ-induced Diabetic Albino Rats

Figure 4 shows that there was an increase in the body weights of the rats during the periods of administration with crude ethanol root-extract of *Sphenocentrum jollyanum* at doses of 250, 500 and 1000 mg/kg body weights and 250 mg/kg body weight of ethylacetate and methanol root fractions. There was no significant difference ($p>0.05$) on twenty one days of treatment when STZ-induced diabetic albino rats treated with crude ethanol root-extract at doses of 250, 500 and 1000 mg/kg body weights and 250 mg/kg body weight of ethylacetate and methanol root fractions were compared with the negative and standard control.

<Insert Figure 4>

DISCUSSION

Diabetes mellitus (DM) is a metabolic disease that results from a defect in insulin action, insulin secretion or both [12]. Insulin deficiency leads to chronic hyperglycemia with disturbances in fat, carbohydrate and protein metabolism [1,16,17,18].

LD_{50} of the crude ethanol root extract of *Sphenocentrum jollyanum*

The LD_{50} of the crude ethanol root extract of *Sphenocentrum jollyanum* in this study is greater than 5000 mg/kg body weight (Table 1) meaning that the crude ethanol root extract is not toxic at this dose. Research and endeavours are geared towards discovery of new therapeutic agents or newer and richer sources of known drugs of natural origin, and the basic goal of such drug discovery efforts always hinges on developing new products with enhanced therapeutic benefits, that is, higher efficacy and low toxicity profile [19,20,21,22,23,24]. While it is not so important to calculate the LD_{50} exactly for substances that are so highly toxic at 1mg/kg body weight, LD_{50} values greater than 5,000mg/kg body weight are of no practical interest because such substances have low-lethality at doses they are likely to be consumed [24]. The results of this study reveal that the extract has LD_{50}values greater than 5,000mg/kg body weight which indicates that they have low-lethality at doses they are likely to be consumed. This is in agreement with the findings of [10], that administered aqueous ethanol root extract of *Sphenocentrum jollyanum* to rabbits and established that the extract administered orally up to the dose of 8000 mg/kg body weight produced no mortality.

Effect of crude ethanol root-extract and fractions of *Sphenocentrum jollyanum* on glucose level in STZ-induced diabetic albino rats

The treatment of STZ-induced diabetic albino rats with ethylacetate and methanol fractions of *Sphenocentrum jollyanum* at doses of 100, 150, 200, 250 and 300 mg/kg body weight significantly ($p<0.05$) decreased the blood glucose level in dose and time dependent manner as shown in figures 1 and 2. Treatment with crude ethanol root-extract at doses of 250, 500 and 1000 mg/kg body weights and 250 mg/kg body weight of ethylacetate and methanol root fractions significantly ($p<0.05$) decreased the blood glucose level in dose and time dependent manner as shown in figure 3. The result equally showed that on day 21 of treatment, the blood glucose level of rats treated with 500 and 1000 mg/kg body weights of ethanol crude root extract and 250mg/kg body weight of ethylacetate and methanol fractions decreased significantly ($p<0.05$) when compared with the positive control rats. But the effect on the standard control was quite similar to that of the crude extract and fractions on day 21 of treatment. The result also showed that there was no significant ($p>0.05$) changes in the glucose level across the groups after the twenty one days of treatment with both 1000 mg/kg body weight of the extract and 250 mg/kg body weight of the fractions as shown in figure 3.

The treatment of STZ-induced diabetic albino rats with the crude ethanol root extract and fractions of *Sphenocentrum jollyanum* significantly decreased ($P<0.05$) the blood glucose levels in rats and this in agreement with the earlier work of [10] who administered aqueous root extract of *Sphenocentrum jollyanum* to alloxan-induced diabetic rabbit and showed that *Sphenocentrum jollyanum* significantly decreased ($p< 0.05$) the blood glucose level from day 3 of daily treatment with the extract. Medicinal plants such as *Sphenocentrum jollyanum and Moringa oleifera* are among the numerous plants adjuncts used in the treatment of diabetes in Africa [25,26,27,28,29]. This is because of its phytochemical constituents which may have some roles in reducing the blood glucose levels [30,31]. Herbal medicines may exert their antidiabetic activity by different modes of action, like acting on insulin secreting pancreatic beta-cells or modifying glucose utilization either by altering the viscosity of gastrointestinal contents, delaying gastric emptying, or delaying glucose absorption from the gut by reducing digestion of polysaccharide (starch) after food intake [32,33,34,35,36].

The return to baseline blood glucose (glycaemia) after treatment with 250 mg/kg body weight of the ethylacetate and methanol fractions was indicative of an enhanced glucose utilization triggered by insulin

production from the beta cells. The fractions showed effective glycaemic control by decreasing the blood glucose concentration. Also the treatment of STZ-induced diabetic rats with the crude ethanol root extract decreased the blood glucose (glycemia) in a dose dependent manner and this appears consistent with the work of [10] who administered ethanol leaf extract of *Sphenocentrum jollyanum* to normal and alloxan-induced diabetic rabbits and showed that the extract exerted significant blood glucose reduction in a dose dependent manner from day 3 of continuous oral administration. Furthermore, previous authors have reported the blood glucose reducing capacity of other medicinal plants like *Allium sativum* [37], *Ageratum conyzoides* [38], *Moringa oleifera* [31], *Bulcholzia coriacea* [39] in STZ and alloxan-induced diabetic rats. WHO asserts that prevention of diabetes and its complications is not only a major challenge for the future, but essential if the health for everyone is to be targeted [40,41,42]. There is still insufficient evidence to draw definite conclusions about the efficacy of medicinal plants, herbs and supplements for diabetes, however, they appear to be generally safe from side effects that can be seen in orthodox medicine [40]. Medicinal plants contain bioactive components that may exert hypoglycaemic effects by reducing insulin resistance, increasing release and decreasing glucagon secretion, slowing the digestion and absorption of carbohydrates or by decreasing hepatic glucose production [28]. *Sphenocentrum jollyanum* also contains flavonoids, terpenoids, glycosides and alkaloids, as its bioactive compounds which elicit their anti-diabetic effect by causing an increase in insulin output or by inhibition of the intestinal absorption of glucose or to the facilitation of metabolites in insulin dependent processes [43, 44]. It could also be achieved through increasing glucose transport (decreasing glycolysis and glucose oxidation in adipose tissue) [22].

Glibenclamide, like the plant extracts also showed significant hypoglycemic activity in the diabetic group of animals. The present findings appear to be in consonance with earlier suggestion of [30] that sulphonylureas such as glibenclamide have extrapancreatic hypoglycemic mechanism of action secondary to their causing insulin secretion and the attendant glucose uptake into and utilization by the tissues. The results from this research show that the extract and fractions at the concentrations of 1000 mg/kg body weight and 250mg/kg body weight of the fractions respectively had similar effects when compared with the standard drug glibenclamide and this is consistent with the results obtained by [12,38].

Effect of crude ethanol root-extract and fractions of *Sphenocentrum jollyanum* on body weight in STZ-induced diabetic albino rats

Figure 4 shows that there was an increase in the body weights of the rats during the periods of administration with crude ethanol root-extract of *Sphenocentrum jollyanum* at doses of 250, 500 and 1000 mg/kg body weights and 250 mg/kg body weight of ethylacetate and methanol root fractions. There was no significant difference ($p>0.05$) on twenty one days of treatment when STZ-induced diabetic albino rats treated with crude ethanol root-extract at doses of 250, 500 and 1000 mg/kg body weights and 250 mg/kg body weight of ethylacetate and methanol root fractions were compared with the negative and standard control.

The treatment of STZ-induced diabetic albino rats with the crude ethanol root extract and fractions of *Sphenocentrum jollyanum* significantly increased the body weights ($P<0.05$) of the rats during the periods of administration when compared to the body weights before induction. This is consistent with the works of [25,43,45] that reported an increase in body weights of diabetic rats treated with ethanol extract of *Ginkgo biloba, Momordica charantia* and *Pterocarpus santalinoides*, respectively. These increases however give the impression that the extract and fractions were generally well tolerated and did not lead to a drastic reduction in food consumption, instead it caused an increase in appetite in the rats, leading to more food consumption and giving credence to the claim that some plant extracts increase

appetite [25]. The findings of this study are also in agreement with the findings of [19] with Nature Cure bitters they worked with. The extract and fractions did not possibly cause any drastic alterations in the carbohydrate, protein or fat metabolism in STZ-induced diabetic rats in such a way as to prevent weight gain expected of animals that are continually supplied with food and water *ad libitum* [19].

CONCLUSION: Crude ethanol root extract, methanol and ethylacetate fractions of *Sphenocentrum jollyanum* reduced the hyperglycemia and boosted body weight of streptozotocin-induced diabetic rats. The ability of the extract and fractions to lower the glucose levels suggest that they possess antidiabetic property which may be due to their chemical constituents. The extract and fractions may therefore be useful in the management and treatment of diabetes and related disorders.

REFERENCES

1. Skyler, J. S. (2004). Diabetes mellitus: pathogenesis and treatment strategies. *Journal of Medical Chemistry,* **47:** 4311-4115.
2. Ivorra, M. D., Paya, M. and Villar, A. (1988). Hypoglycaemic and Insulin release effects of tormentic acid: A new hypoglycaemic natural product. *Plant Medicine,* **54:** 282-286.
3. Zurina, H., Mun, F. Y., Mariam, A. and Ahmad, P. (2010) Mechanism of action of *Gynura procubens* water extract in streptozotocin-induced diabetic Rats. *Molecules*, **15:** 9008-9023.
4. Ugwu Okechukwu P.C. and Amasiorah V.I. (2020) The effects of the crude ethanol root extract and fractions of *Sphenocentrum jollyanum* on hematological indices and glycosylated haemoglobin of streptozotocin-induced diabetic albino rats. *INOSR Scientific Research*,6(1): 61-74.
5. Ibironke, A. A. and Olusola, O. O. (2013). Chemical composition of ten medicinal plant seeds from South-West Nigeria. *1ISTE Advances in Life Science and Technology*, **10:** 25-32.
6. Nia, R., Paper, D. H., Essien, E. E., Lyadi, K. C., Bassey, A. I. L., Anti, A. B. and Franz, G. (2004). Evaluation of the anti-oxidant and anti-angiogenic effects *of Sphenocentrum jollyanum* Pierre. *African Journal of Biomedical Research,* **7:** 129-132.
7. Woode, E., Amidu, N., William, K. B. A., Ansah, C. and Duwiejva, M. (2009). Anxiogenic-like effects of root extract of Sphenocentrum jollyanum Pierre in Murine behavioral models. Journal of Pharmacology and Toxicology,4(3): 91-106.
8. Owiredu, W. K. B. A., Nafiv, A., Amissah, F. and Woode, E. (2007). The effects of the ethanolic extract of root *of Sphenocentrum jollyanum* Pierre on sexual behaviour-and hormonal levels in rodents. *Journal of Science and Technology,* **27**(2): 9-20.
9. Ekpono, E. U., Aja, P. M., Ibiam, U. A., Alum, E. U. and U. E. Ekpono. (2019). Ethanol Root-extract of *Sphenocentrum jollyanum* Restored Altered Haematological Markers in *Plasmodium berghei*-infected Mice. *Earthline Journal of Chemical Sciences*,**2** (2):189203. https://doi.org/10.34198/ejcs.2219.189-203.
10. Mbaka, G. O., Adeyemi,O. O., Anunobi, C. C., Noronha, C. C. and Okanlawon, O. A. (2008). Anti-hyperglycaemic effects of ethanol leaf extract of *Sphenocentrum jollyanum* in normal and alloxan-induced diabetic rabbits. *Global Journal of Pharmacology*, **2** (3): 46-51.
11. Akintobi, O. A., Adejuwon, A. O., Bamkefa, B. A.,Daniels, O. V. C and Ojo, V. O. (2013). Antimicrobial potency of *Sphenocentrum jollyanum* on some human pathogenic bacteria. *Academia Arena,* **5**(5): 1 -7.
12. Alese, M. O., Adewole, O. S., Ijomone, O. M., Ajayi, S. A. and Alese, O. O. (2014). Hypoglycemic and hypolipidemic activities of rnethanolic extract of *Sphenocentrum jollyanum* on streptozotocin-induced diabetic wistar rats, *European Journal of Medicinal Plants,* **4**(3):353-364.
13. Muko, K. N., Ohiri, P. C. and Ezegwu, C. O. (1998). Antipryetic and analgesic activities of *Sphenocentrum jollyanum. Nigerian Journal of Natural Product Medicine,* **2:**52-53.
14. Samuel, F. O., Sinbad, O. O. and Olusoji, O. In vitro anti-inflammatory activities of extrct of the leaves of *Sphenocentrum jollyanum* Pierre. *Journal of applied life sciences international*, 2018; **18**(4): 1-9.

15. Sinbad, O. O., Samuel, F. O., Adewale, A. and Adedoyin, O. Evaluation of Membrane Stabilizing, Proteinase and Lipoxygenase Inhibitory Activities of Ethanol Extract of Root and Stem of *Sphenocentrum jollyanum* Pierre. *Journal of Advances in Biology and Biotechnology*, 2017; **3**(1): 1-8.
16. Szkudelski, T. (2001). The mechanism of alloxan and streptozotocin action in the β-cells of the Rat pancreas. *Physiological Research*, **50:** 536-546.
17. Lenzen, S. (2008). The mechanisms of alloxan and streptozotocin-induced diabetes. *Diabetologia*, **51:** 216-226.
18. Akbarzadeh. A., Norouzian. D., Mehrabi. M. R., Jamshidi, S., Farhangi, A., Allah, A. V., Mofidian. S. M. A. and Lame, B. R. (2007). Induction of diabetes by streptozotocin in rats. *Indian Journal of Clinical Biochemistry*, **22**(2): 60-64.
19. Lorke, D. (1983). A new approach to practical acute toxicity testing. *Archives of Toxicology*, **53:** 275 -280.
20. Rossini, A. A., Like, A. A.,Appel, M. C. and Williams, R. M.(1978). Streptozotocin-induced pancreatic insulitis in mice. Morphologic and physiologic studies. *Laboratory Investigations*, **38:** 86-470.
21. Aniagu, S. O., Nwinyi, F. C., Akumka, D. D., Ajoku, G. A., Dzarma, S., Izebe, K. S., Ditse, M., Nwaneri, P.E.C., Wambebe, C. and Gamaniel, K. (2005). Toxicity studies in rats fed nature cure bitters. *African Journal of Biotechnology*,**4**(1): 72-78.
22. Babayemi, J. O. Dauda, K. T. Nwude D. O. and. Kayode, A. A. A. (2010). Evaluation of the composition and chemistry of ash and potash from various plant materials. A review. *Journal of Applied Sciences*, **10:** 1820-1824.
23. Bhandaria, M. R., Jong-Anurakkuna, N. and Honga, G. (2008). α-Glucosidase and α-amylase inhibitory activities of Nepalese medicinal herb *Bergenia ciliate*.*Journal of Food Chemistry*, **106**: 247-252.
24. Clark, F. J., Burgess, R. C., Chapin, J. W. and Lipscomb, W. T. (1985). Role of intramuscular receptors in the awareness of limb position. *Journal of Neurophysiology*, **54:** 1529-1540.
25. Daye, C., Bin, L. and Yunhui, L. (2012). Antihperglycemic effect of *Ginkgo biloba* extract in streptozotocin-induced diabetes in rats. *Biomed Research International*, **13:** 1-7.
26. Gamaniel, K.S. (2000). Toxicity from medicinal plants and their products. *NigerianJournal of Natural Product Medicine*,**4:** 04-09.
27. Hoffmann, D. (2000). *Healthy Digestion: A Natural Approach to Relieving Indigestion, Gas, Heartburn, Constipation, Colitis & More*, (1st edition). Storey Publishing, LLC, Massachusetts. Pp. 1- 123.
28. Iwu, M. M. (1993). Handbook of African Medicinal Plants. CRC Press Incorporation. Pp. 239-245.
29. Jack, D. (2003). Overview of the anti-diabetic agents. *Endocrine Pharmacotherapy Module, Spring*,**7:** 1-33.
30. Jackson, J. E. and Bressler, R. (1981). Clinical pharmacology of sulphonylurea hypoglycemic agents. *Drugs*, **22:** 211-245

31. Aja, P.M., Igwenyi, I.O., Ugwu, O.P.C., Orji, O.U. and Alum, E.U. (2015). Evaluation of Anti-diabetic Effect and Liver Function Indices of Ethanol Extracts of *Moringa oleifera* and *Cajanus cajan* Leaves in Alloxan Induced Diabetic Albino Rats. *Global Veterinaria*,14 (3): 439-447.DOI: 10.5829/idosi.gv.2015.14.03.93129.
32. Ugwu Okechukwu PC, FC Nwodo Okwesili, E Joshua Parker, Bawa Abubakar, C Ossai Emmanuel, E Odo Christian (2013). Phytochemical and Acute Toxicity Studies of *Moringa oleifera* Ethanol Leaf Extract. *International Journal of Life Science Biotechnology and Pharma Research*,2(2): 66-71.
33. Enechi, O.C., Ugwu, Kenneth K., Ugwu Okechukwu P.C. and Omeh YS (2013). Evaluation of the Antinutrient Levels of Ceiba Pentandra Leaves. *IJRRPAS* 3 (3), 394-400.
34. Afiukwa, C. A., Ogah, O., Ugwu, O. P. C., Oguguo, J. O., Ali, F. U., and Ossai E. C (2013). Nutritional and Antinutritional characterization of two wild Yam species from Abakaliki, Southeast Nigeria. Research Journal of Pharmaceutical, Biological and Chemical Sciences,4(2): 840-848.
35. Edeoga, H.O., Okwu, D.E. and Mbaebie, B.O. (2005). Phytochemical constituents of some Nigerian medicinal plants. *African Journal Biotechnology*,**4**(7):685- 688.
36. Ugwu Okechukwu P.C. and Amasiorah V.I. (2020). The In Vivo Antioxidant Potentials of the Crude Ethanol Root Extract and Fractions of Sphenocentrum jollyanum on Oxidative Stress Indices in Streptozotocin-Induced Diabetic albino rats. IDOSR Journal of Biology, Chemistry and Pharmacy, 5(1): 26-35.
37. Offor, C.E., Ugwu, O.P.C. and Alum, E. U. (2014). The Anti-Diabetic Effect of Ethanol Leaf-Extract of *Allium sativum* on Albino Rats. *International Journal of Pharmacy and Medical Sciences*, 4 (1): 01-03.
38. Agbafor, K.N., Onuoha, S.C., Ominyi, M.C., Orinya, O.F., Ezeani, N. and Alum, E.U. (2015). Antidiabetic, Hypolipidemic and Antiathrogenic Properties of Leaf Extracts of Ageratum conyzoides in Streptozotocin-Induced diabetic rats. *International Journal of Current Microbiology and Applied Sciences*, 4 (11): 816-824.
39. Egwu, C. O., Offor, C. E. and Alum, E. U. (2017). Anti-diabetic effects of *Buchholzia coriacea* ethanol seed Extract and Vildagliptin on Alloxan-induced diabetic albino Rats. *International Journal of Biology, Pharmacy and Allied Sciences (IJBPAS)*, **6 (6):** 1304-1314.
40. World Health Organization (2008). Africa Traditional Medicine. *Medicinal Journal of Australia*, **30**:224-226.
41. Prakash, D., Kumar, P and Kumar, N. (2009). Antioxidant and Hypoglycemic Activity of Some Indian Medicinal Plant, Pharmacologyonline, **3:** 513-521.
42. Priyankar, D., Manas, R. S., Sumedha, R. C, Arab, S., Mousumi, P. S, Biswajit, H and Tapas, K. C. (2015). Assessment of anti-diabetic activity of an ethnopharmacological plant Nerium oleander through alloxan induced diabetes in mice. *Journal of Ethnopharmacology*, **161:** 128-137.
43. Shirin, M., Mondal, D. K. and Shamim, K. M. (2011). Impact of *Momordica charantia* on the body weight in Streptozotocin-induced diabetic rats. *Bangladesh Journal of Anatomy*, **9** (2): 106-109.
44. Weiss, R. B. (1982). Streptozotocin; A review of its pharmacology, efficacy and toxicity. *Cancer Treatment and Reproduction*, **66:** 427-438.

45. Aja, P.M., Ani, O.G., Offor, C.E., Orji, U.O. and Alum, E.U. (2015). Evaluation of Anti-Diabetic Effect and Liver Enzymes Activity of Ethanol Extract of *Pterocarpus santalinoides* in Alloxan Induced Diabetic Albino Rats. *Global Journal of Biotechnology & Biochemistry***10** (2): 77-83.DOI: 10.5829/idosi.gjbb.2015.10.02.93128.

Table 1: Acute toxicity and lethal dose (LD_{50}) assessment of the Crude Ethanol Root Extract of *Sphenocentrum jollyanum*

Group	No of Mice	Dose (mg/kg Body Weight)	Death and Signs of Toxicity
1	4	Control	ND and NST
2	4	10	ND and NST
3	4	100	ND and NST
4	4	1000	ND and NST
5	4	1900	ND and NST
6	4	2600	ND and NST
7	4	5000	ND and ST

Key: ND= No death; NST= No signs of toxicity; ST= Signs of toxicity

Figure legends

Figure 1: Effect of ethylacetate fraction of *Sphenocentrum jollyanum* on glucose levels in STZ-induced diabetic wistar albino rats. Data are shown as Mean ± standard deviation (n=4). Mean values with different alphabet showed significant difference at $p<0.05$.
Key: EAF= Ethylacetate fraction

Figure 2: Effect of methanol fraction of *Sphenocentrum jollyanum* on glucose levels in STZ-induced diabetic wistar albino rats. Data are shown as mean ± standard deviation (n=4). Mean values with different alphabet showed significant difference at $p<0.05$.
Key: MF= methanol fraction

Figure 3: Effect of crude ethanol root-extract and fractions of *Sphenocentrum jollyanum* on glucose levels in STZ-induced diabetic wistar albino rats. Data are shown as mean ± standard deviation (n=4). Mean Values with different alphabet showed significant difference at $p<0.05$.
Key: CERE=Crude ethanol root extract, MF= Methanol fraction and EAF= Ethylacetate fraction, BI= before induction, 3 DAYS AI= three days after induction, DAY 14 T= day fourteen of treatment, DAY21T =day twenty one of treatment.

Figure 4: Effect of crude ethanol root-extract and fractions of *Sphenocentrum jollyanum* on body weight in STZ induced diabetic wistar albino rats. Data are shown as mean ± standard deviation (n=4). Mean values with different alphabet showed significance difference at $p<0.05$.
Key: CERE=Crude ethanol root extract, MF= Methanol fraction and EAF Ethylacetate fraction, WBI= Weight before induction, WA2WT=Weight after two weeks of treatment and WA3WT=Weight after three weeks of treatment.

Figure 1

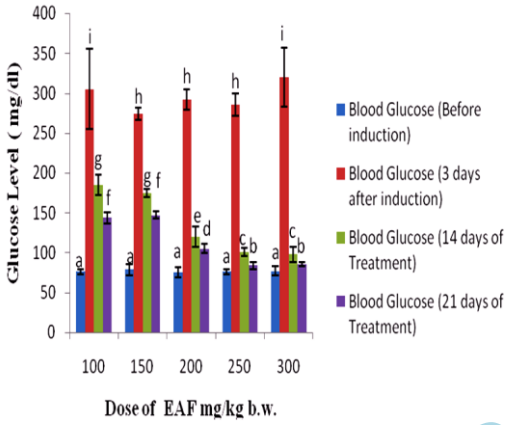

Figure 2

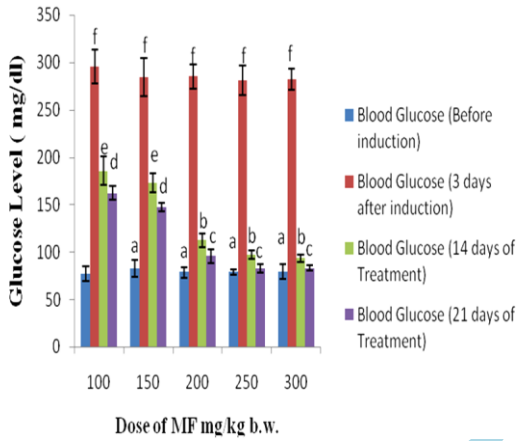

Figure 3

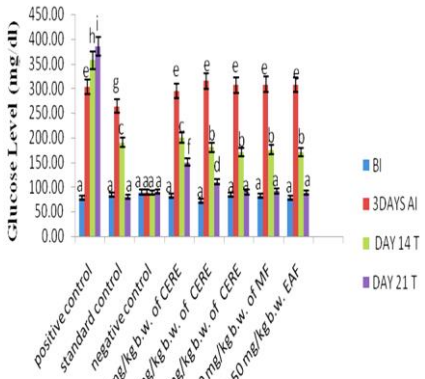

Figure 4