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Studies on Effects of *Gmelina arborea* Bark Extract in Blood Glucose Level and Pancreatic Histology

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Abstract:- Studies on effects of Gmelina arborea bark aqueous extract in glucose level and pancreatic histology of experimentally induced albino rats were undertaken. Twenty albino rats were used for this study. The animals were grouped into five, containing four each. Normal control(group I) administered 0.4ml/Kg.bw. distilled water, group II(0.4ml/Kg.bw. alloxan monohydrate) group III(0.4ml./Kg.bw of alloxan + 0.5ml/Kg.bw. glibenclamide, group IV (0.4ml/Kg.bw. alloxan + 200mg/Kg.bw. Gmelina arborea stem bark aqueous extract) and group V received 0.4ml/Kg.bw. alloxan + 400mg/Kg.bw. extract. The treatments were orally administered daily to the albino rats for twenty-one days. The fasting blood glucose levels were measured on day 0, 1,7,14 and 21. On 21st day after the blood glucose measurement, all the animals were sacrificed and their pancreas were removed and processed for histological examination. The results revealed reduction in the glucose levels of the animals treated with aqueous extract of Gmelina arborea stem bark. The treatment with the aqueous extract ameliorated the weight loss caused by the diabetic. The histology of the pancreas revealed areas of regeneration changes in group IV and V when compared with group II animals which showed areas of fibrosis of the tissue stroma viscurity. This study suggests that aqueous extract of Gmelina arborea stem back may ameliorate diabetic condition and other related disorders.

Keywords:- *Antidiabetic , Hyperglycemia, Glucose level, Pancreas, Gmelina arborea, Alloxan.*

I. INTRODUCTION

Gmelina arborea commonly known as beechwood is a well-known medicinal plant in an ancient India system of medicine. The roots, leaves, flowers, fruits and bark are used for treating different ailments in traditional medicine. The literature suggests use of the plant in treatment of scorpion sting, snake-bites, (Nadkarni, 2000), and diabetes (Khan and Khanum, 2005). The plant is anthelmintic and used for treating hallucinations, excess thirst, piles, abdominal pains, burning sensations, and fever, (Kirtikar and Basu, 1999).Crude extracts of the plants are reported to possess wound healing properties (Shirwaikar, *et al.* 2003), antidiarrheal activity (Agunu, *et al*. 2005), antioxidant activity (Sinha *et al*. 2006), antidiabetic activity (Kulkarni and Addepalli, 2011), and antiulcer activity (Giri, *et al.* 2009).

Diabetes mellitus known as diabetes is a disorder that alters the metabolism of carbohydrates, fats, and proteins. The disorder results from the shortage of insulin or lack of it or reduced sensitivity of the tissues to insulin (Piero, *et al.* 2015). About 25% of the world's population according to a survey is reported to have *Diabetes mellitus.* The prevalence of diabetes for all ages groups worldwide is on the increase and has been projected as the world's main disorder and killer in the next 25 years (Edwin, *et al.* 2006).The number of people suffering from diabetes worldwide is increasing at an alarming rate(Wild, *et al.* 2004).This is because none of the antidiabetic drugs could give a long term glycemic control without causing adverse side effects(Singh *et al*.2007). Food provides protection because they contain beneficial phytochemicals which decrease the incidences of diabetes and metabolic syndrome (Sciechitano, *et al.* 2014).

Medicinal plants that are effective in controlling plasma glucose level with minimal side effects are commonly used in under developed countries as alternative therapy. In Africa, hundreds of plants are used traditionally for the management and control of *Diabetes mellitus*. Unfortunately, only a few of such medicinal plants have been scientifically validated (Tanko, *et al. 2*007).Therefore the aim of this study was to investigate the effect of *Pmelina arborea* stem bark aqueous extract on blood glucose level and pancreatic histology of albino rats.

II. MATERIALS AND METHODS

➢ *Plant Material*

Gmelina arborea stem bark harvested in the farmland of Federal Polytechnic Nekede, Owerri, was identified by a botanist.

➢ *Preparation of Gmelina arborea Extract*

The bark was washed with distilled water, dried at room temperature and was ground using electric blender.

Hundred gram (100g) of the powder was macerated with 1000ml distilled water, for 6 hours with occasional shaking. After 6 hours, filtration funnel and whatman No 1 filter paper were use and the filtrate collected and stored in airtight bottle at -20^{o}C.

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❖ *Experimental Animals and Design*

Twenty (20) albino rats of 6-8 weeks old and weight of 90-150g were used for the study. Animal feed and water was used to feed the albino rats and they were kept in well ventilated animal house condition for acclimatization period of two weeks before the commencement of the experiment.

The albino rats were fasted overnight. This made them to become hypoglycemic, alloxan monohydrate was used to induce diabetes *mellitus* by injecting single dose (125mg/kg) of freshly prepared 5% solution of alloxan monohydtate in 0.9% sodium chloride (Normal saline) intraperitoneally.After 48 hours, glucose level was determined .Those albino rats showing blood glucose level of 126mg/dl or more than were selected for the study. The animals were randomly selected into five(5) groups containing four(4) each. The aqueous extract of *Gmelina arborea* bark and Glibenclamide were administered consecutively for 21 days. The treatment protocol is as follows:

Groups	Number of albino rats	Treatments
Group I (Normal control)	4	Normal control rats administered 0.4ml/Kg distilled water
Group II (Diabetic control)	4	Diabetic rats received 0.4ml/Kg alloxan monohydrate
Group III	4	Diabetic albino rats received 0.4ml/Kg of alloxan monohydate + 0.5mg/Kg Gibenclamide
Group IV	4	Diabetic rats received 0.4ml/Kg of alloxan monohydrate + 200mg/Kg *Gmelina arborea* extract
Group V	4	Diabetic rats received 0.4ml/Kg of alloxan monohydrate + 400mg/Kg *Gmelina arborea* extract daily.

Table 1

➢ *Treatment and Termination of Experiment*

The blood glucose level was determined by using glucometer on day 0, 1, 7, 14 and 21 by tail blood withdrawal techniques, after the overnight fasting. On 21st day all the animals were sacrificed by cervical dislocation after the last blood sampling and the pancreas were sent for histopathological examination.

➢ *Histopathological Examination*

Each of the pancreas was fixed in 10% formalin and undergone a standard tissue processing for histopathological examination and thereafter a careful sections of 4µ (four micron) thickness, were cut from the tissue and stained with Haematoxylin and Eosin stains.

➢ *Statistical Analysis*

The results were analyzed for statistical significance using one-way ANOVA. The p-value > value 0.05 was considered not significant. The data were expressed as (Mean ± SD) n= 4.

III. RESULTS

The glucose levels of the experimental animals were determined. At Day 0 all albino rats used in this study have plasma blood sugar level ranges from 57.25mg/dl to 90.25mg/dl for rats in the normal and diabetic control group, respectively. This shows that all animals used in this study are free from diabetes characterized by high blood sugar level (Table 2).

Group	Treatments	Mean ± SD Glucose Level (mg/dl) at Day 0
I(Control)	0.4ml/Kg distilled water	57.25±16.32
II(Diabetic control)	0.4ml/Kg Alloxan monohydrate	90.25±8.96
III	0.4ml/Kg Alloxan monohydrate + Glibenclamide (0.5mg/Kg)	86.75±11.15
IV	0.4ml/Kg Alloxan monohydrate +*Gmelina arborea* (200mg/Kg)	79.75±9.43
V	0.4ml/Kg Alloxan monohydrate +*Gmelina arborea* (400mg/Kg)	80.25±12.92

Table 2

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Group	Treatments	Mean ± SD Glucose Level (mg/dl) at Day 1
I(Control)	0.4ml/Kg distilled water	60.25±11.03[b]
II(diabetic control)	0.4ml/Kg Alloxan monohydrate	323.00±59.30[a]
III	0.4ml/Kg Alloxan monohydrate + Glibenclamide (0.5mg/Kg)	316.75±122.53[a]
IV	0.4ml/Kg Alloxan monohydrate + *Gmelina arborea* (200mg/Kg)	358.75±101.70[a]
V	0.4ml/Kg Alloxan monohydrate + *Gmelina arborea* (400mg/Kg)	369.50±125.55[a]

Table 3

a, b-superscripts indicate significant difference ($p<0.05$)

Group	Treatments	Mean ± SD Glucose Level (mg/dl) at Day 7
I (control)	0.4ml/Kg distilled water	63.00±6.98[b]
II(Diabetic control)	0.4ml/Kg Alloxan monohydrate	351.75±109.58[a]
III	0.4ml/kg Alloxan monohydrate + Glibenclamide (0.5mg/Kg)	239.50±146.56[ab]
IV	0.4ml/Kg Alloxan monohydrate + *Gmelina arborea* (200mg/Kg)	316.00±116.87[a]
V	0.4ml/Kg Alloxan monohydrate + *Gmelina arborea* (400mg/Kg)	264.00±156.58[a]

Table 4

a, b, ab-superscripts indicate significant difference ($p<0.05$)

Group	Treatments	Mean ± SD Glucose Level (mg/dl) at Day 14
I(Control)	0.4ml/Kg distilled water	67.50±8.23[c]
II(Diabetic control)	0.4ml/Kg Alloxan monohydrate	324.75±58.83[a]
III	0.4ml/Kg Alloxan monohydrate + Glibenclamide (0.5mg/Kg)	177.50±99.45[bc]
IV	0.4ml/Kg Alloxan monohydrate *Gmelina arborea* (200 mg/Kg)	259.00±104.63[ab]
V	0.4ml/Kg Alloxan monohydrate *Gmelina arborea* (400 mg/Kg)	148.50±48.91[bc]

Table 5

a, b, c-superscripts indicate significant difference ($p<0.05$)

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Group	Treatments	Mean ± SD Glucose Level (mg/dl) at Day 21
I(Control)	0.4ml/Kg distilled water	64.50±6.60[b]
II(Diabetic control)	0.4ml/Kg Alloxan monohydrate	321.75±58.52[a]
III	0.4ml/Kg Alloxan monohydrate + Glibenclamide (0.5mg/Kg)	91.25±7.18[b]
IV	0.4ml/Kg Alloxan monohydrate *Gmelina arborea* (200 mg/Kg)	79.50±25.05[b]
V	0.4ml/Kg Alloxan monohydrate + *Gmelina arborea* (400 mg/Kg)	60.75±23.26[b]

Table 6

a, b,-superscripts indicate significant difference ($p<0.05$)

Group(s)	Treatments	Mean ± SD weight gain/loss
I	Distilled water(0.4ml/Kg)	25.35±2.67[a]
II	Alloxan (0.4ml/Kg)	-29.9725±1.20[d]
III	Alloxan(0.4ml/Kg)+Glibenclamide (0.5mg/Kg)	6.6025±2.61[c]
IV	Alloxan(0.4ml/Kg)+*Gmelina arborea* (200 mg/Kg)	6.9775±4.21[c]
V	Alloxan(0.4ml/Kg)+*Gmelina arborea* (400 mg/Kg)	19.4275±1.77[b]

Table 7

a, b, c, d, superscripts indicate significant difference ($p<0.05$)
+ve value means weight gain
-ve value means weight loss

The rats in the diabetic control group recorded weight loss. The highest increase in weight was recorded in rats given *G arborea* at 400mg/Kg body weight followed by the rats in group IV which shared similar effect on weight with the glibenclamide rats.

❖ *Histopathological examination of pancreas*

Histopathological results as shown in Plate1, the pancreas of the albino rat in the normal control appears normal with no fibrosis.

Below Plates show the efficacy of aqueous extract of *G. arborea* in reversing the fibrosis: In Plate 3 there is appearance of fibrosis, while in Plate 4 after administration of glibenclamide drug the rats recover from the tissue stroma, the same results were observed in the aqueous extract of *G. arborea* as shown in Plate 4 and 5.

➢ *Light microscopic observation of thin section of the Pancreas after H and E stain. A (X 160)*

Plate 1:- Photomicrograph of Pancreas section of Normal control

- *Light microscopic observation of thin section of the Pancreas after H and E stain.B (X 160)*

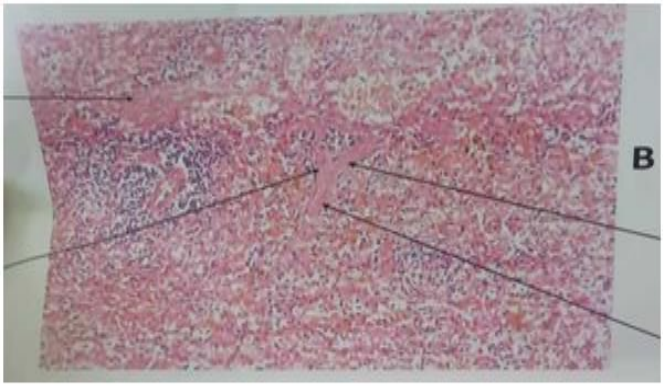

Plate 2:- Photomicrograph of Pancreas section of Diabetic control B, Arrows showing areas of fibrosis of the tissue stroma vascurity.

- *Light microscopic observation of thin section of the Pancreas after H and E stain.C (X 160)*

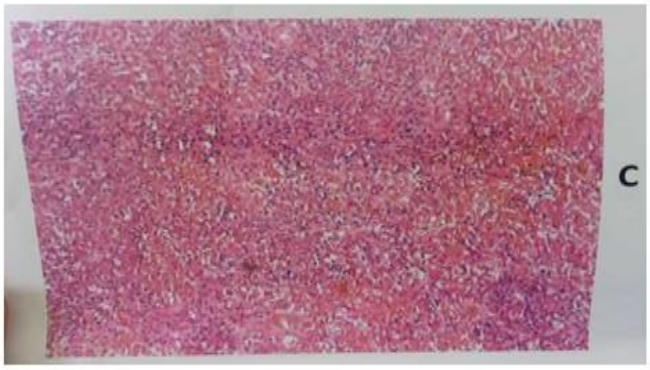

Plate 3:- Photomicrograph of Pancreas section of drug control (c), appears normal

- *Light microscopic observation of thin section of the Pancreas after H and E stain.D (X 160)*

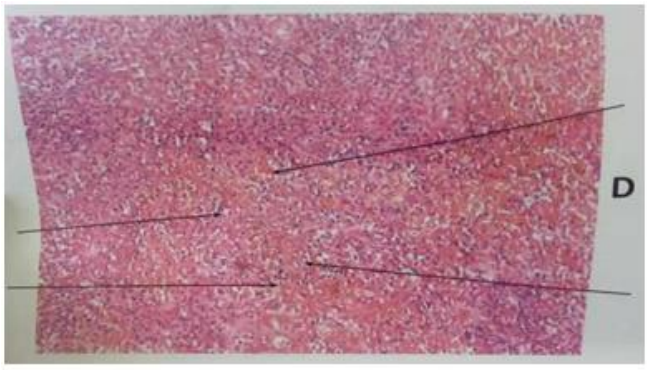

Plate 4:- Photomicrograph of Pancreas section after treatment with aqueous extract of *G. arborea* at 200mg/Kg. Arrows showing areas of regeneration changes. Therefore there is recovering of tissue stroma

- *Light microscopic observation of thin section of the Pancreas after H and E stain.E (X 160)*

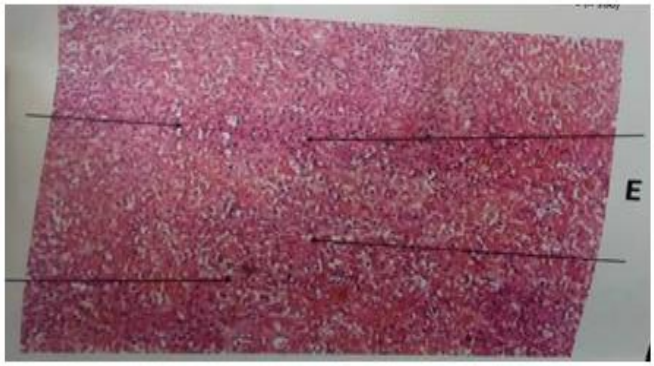

Plate 5:- Photomicrograph of Pancreas section after treatment with aqueous extract of *G. arborea* at 400mg/Kg. Arrows showing areas of regeneration changes. Therefore there is recovering of tissue stroma

IV. DISCUSSION

In this study, effect of the aqueous extract of *Gmelina arborea* stem bark in blood glucose level and pancreatic histology of alloxan induced albino rats was investigated. After successful determination of the blood glucose level in albino rats, it was discovered that the effect of the aqueous extract of *G. arborea* were gradually noticed from Day 1 to Day 21 when its effect was greatly observed. This is in agreement with the study by Kulkarni and Veerranjaneyulu (2013).Who reported anti-hyperglycemic activity of the extract.

Glibenclamide was used to compare the efficacy of aqueous extract of stem bark of *G. arborea,* which was related to the study of Kulkarni and Veerranjaneyulu (2013) but at a dose 0f 0.6mg/Kg body weight daily rather than 0.5mg/Kg daily used in this study.

However, the information on effect of the extract on weight revealed a significant ($p<0.05$) weight loss in diabetic control rats. The dosage of aqueous extract shows significant effect on weight gain as the rat in group V recorded the highest weight gain. This is also in agreement with Kulkarni and Veerranjaneyulu (2013). Who reported reduction in body weight loss with the treatment with extract of *G. arborea.* This work is in a way corroborates the work of Kumaresan *et al*. 2014. They reported that the plant extract was nontoxic and possessed antidiabetic activity.

Histopathological study on the pancreas revealed fibrosis on its stroma tissue of diabetic control group, the case which was corrected by glibenclamide at a dose of 0.5mg/Kg and extract at both 200 and 400mg/Kg. This is in agreement with the submission of John's Hopkins Fibrosis Centre (2018).

V. CONCLUSION

In conclusion, this study has shown that the *Gmelina arborea* extract competed well in lowering blood sugar level with standard anti-diabetic drug (glibenclamide at 0.5mg/Kg).The extract improved weight loss that accompanied diabetic induction. The extract also reversed the fibrosis of tissue stroma.

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