Evaluation of Renoprotective Potential of Plantain (*Musa parasidiaca***) Stem Juice on Rat Model Diabetes**

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Abstract— The prevalence of diabetes mellitus in the world is increasing. Hyperglycemia in diabetes mellitus can increase the risk of complications in various organs, one of which is chronic renal failure because symptoms of type 2 diabetes, unlike type 1 diabetes, do not occur, the screening methods for identifying kidney involvement is very important. In chronic renal failure, a decline in kidney function is chronic, resulting in increase of the levels of blood urea nitrogen (BUN) and creatinine in the blood. A level of BUN and creatinine serum is use as a parameter of kidney function tests. Plant-derived medicinal products have attracted the attention of scientists around the world for many years due to their minimum side effects and positive effects on human health hence the choice of plant used in the present study. 35 healthy Albino (Wister strain) were obtained and grouped into seven. The results obtained from sub-diabetic and diabetic rats both treated with 250mg/kg/bw and 500mg/kg/bw respectively when compared with diabetic control, plantain stem juice shows a lowering effect on the blood creatinine and BUN concentration. The effect of the stem juice of *Musa paradisiaca* suggests that it can be useful in controlling creatinine and BUN concentration in diabetics and will serve as a better alternative to synthetic drugs.

Keywords— Diabetes; BUN; Creatinine; Alloxan; Musa parasidiaca; Rats.

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I. INTRODUCTION

Diabetes mellitus (MD) affects approximately 4% of the population worldwide and is expected to increase by 4.5% in 2025. Diabetes mellitus type 1 (DM1) is the leading cause of renal failure, causing about 45% of new cases each year. Even when diabetes is controlled and blood sugar levels are maintained, the kidneys may be damaged or may fail. Diabetic nephropathy affects approximately one third of diabetes patients and is the leading cause of end-stage renal disease in many countries [1]. In chronic renal failure, a decline in kidney function is chronic, resulting in glomerular filtration rate falling by more than 50% and will increase the levels of Blood Urea Nitrogen (BUN) and Creatinine in the blood. Levels of BUN and creatinine serum are used as a parameter of kidney function tests [2].

Animal experimentation has a long history of contributions to the fields of diabetes and metabolic disease research. Currently, most experiments are performed in rodents. Several toxins, including streptozotocin and alloxan, induce type 1 diabetes mellitus (DM1) [3]. Plant-derived medicinal products have attracted the attention of scientists around the world for many years due to their minimum side effects and positive effects on human health [4]. Today, medicinal plants play a great role in human health services worldwide. Many people in the modern world are turning to herbal medicine. For example, in USA about

25% of all prescriptions dispensed in public pharmacies in 1973 contained drugs extracted from higher plants and about 64% of the total global populations remain dependent on traditional medicine for their healthcare needs [5]. Medicinal plants are frequently used in traditional medicine to treat different diseases in different areas of the world [6]. This indigenous knowledge, passed down from generation to generation in various parts of the world, has significantly contributed to the development of different traditional systems of medicine [7] as well as helped in exploration of different medicinal plants to find the scientific basis of their traditional uses [8]. Traditional practice medicinal plants are used in many countries to control diabetes mellitus. The hypoglycemic actions of these medicinal plants are being studied. Plant drugs are frequently considered to be less toxic and freer from side effects than synthetic ones [9]. Nigerian medicinal plants are known to contain a large variety of phytochemical components in the search for potential new drugs against the illness [10].

Inadequate production and knowledge of the medicinal uses of plantain stem juice and cultural attitudes toward certain medicinal plants such as plantain stem juice has resulted to underutilization in our local society. This study is aimed at evaluating the renoprotective potential of *Musa paradisiaca* stem juice on induced diabetic albino rats grown in their natural environment in Idah, Kogi State.

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Taxonomy Classification of Plantain Plant:

Kingdom: Plantae Division: Magnoliophyta

Class: Liliopsida Order Zingiberales Family Musaceae Genus Musa

Species: Musa paradisiaca

Description of Plantain:

Musa paradisiaca is a herbaceous plant (up to 9 m long) with a robust tree like pseudo stem, a crown of large elongated oval deep-green leaves (up to 365 cm in length and 61cm in width), with a prominent mid rib each plant produces a single inflorescence like drooping spike, and large bracts opening in succession, ovate, 15-20 cm long, concave, dark red in color and somewhat fleshy. Fruits are oblong, fleshy, 5-7cm long in wild form and longer in the cultivated varieties. Musa paradisiaca, commonly known as the plantain, is an herbaceous plant extensively cultivated in tropical and subtropical regions. Owing to its worldwide consumption, the plant is widely distributed in the southern part of Nigeria [11].

Medicinal Importance of Plantain:

The plantain is of great economic importance. It should be noted that all parts of the plantain plant, including leaves, peels, root and stems have been used in folk medicine to treat various disorders such as diarrhea, dysentery, intestinal colitis, inflammation, pain and snakebite. Furthermore, a number of pharmacological activities have been reported not only for Musa paradisiaca, but also for wild species, represented mainly by Musa acuminate [12]. In the last decade, special attention has been paid to the potential of extracts derived from different parts of the plantain plant in supporting the management of both type one [13] and type two diabetes mellitus [14], a metabolic disease whose incidence has been growing significantly in the Western world. Since diabetes has a multifactorial etiology, various underlying mechanisms have been investigated. In addition to the ability to inhibit carbohydrate digesting enzymes (glycosidase amylase), the potential of plantain phytochemicals to affect, e.g., glucose adsorption and/or antioxidant activity have been most frequently described. However, it is worth noting that the concentrations of secondary metabolites contained in plantain plants, and, consequently, the bioactivities of obtained extracts may vary depending not only on respective species, but also due to multiple environmental factors and industries [15].

II. RELATED WORK

The green fruit of M. paradisiaca has been reported to have hypoglycemic effect due to stimulation of insulin production and glucose utilization [16]. Its high potassium (K) and sodium (Na) content has been correlated with the glycemic effect [17]. The anti-hyperglycemic effect of the hydromethanolic extract of M. paradisiaca root has been found significant [18]. It also controls lipid peroxidation in diabetes [19].

III. METHODOLOGY

Sample Collection and Identification: Musa paradisiaca (Plantain) which is grown at Okenya in Idah Local Government Area of Kogi State was used in the present study. The plant materials were procured from the local growers. Identification and authentication of the stem alongside the fruits and leaves was carried out by Mr. C.J. Onyeukwu, a Plant Taxonomist, in the Department of Plant Science and Technology, University of Nigeria Nsukka, Nigeria. 35 healthy Albino (Wister strain) rats of 3 months of age were obtained from the animal house of University of Nigeria Nsukka, Enugu State for this experiment and were grouped into seven.

Study Design: Rats were grouped into seven. Experimental animals were fed with normal feeds (marsh grower). Induction of diabetes was done with alloxan monohydrate. On 16th day of experiment, all the animals were sacrificed and blood collection was done via cardiac puncture. The blood samples of both diabetic and subdiabetic rats collected were analyzed on the same day of collection and serum was separated by centrifugation at 3000rpm for 5 min for the assay of the blood urea nitrogen and creatinine concentration in the blood samples, using the Cobas 111 instrument.

Study Location and Duration: This research work was carried out at the Federal Polytechnic Idah, Kogi State, in the Department of Science and Laboratory Technology from December 2018 to December 2019.

Sample Size: 35 healthy Albino (Wister strain) rats of 3 months of age were used in the present study. Out of 5mL of plantain stem juice collected, exactly 0.604mL of the juice was administered to the experimental animals at the end of the study.

Dosage Determination

5mL of the stem juice was placed in a porcelain dish. This was then transferred to an oven and dried at 105°C till constant weight. This was carried out in triplicate. Their average dry weight was calculated. The residue obtained on evaporating 5mL of stem juice to dryness was 0.09g. From ethno medicine, 30mL is taken by an average 70kg man which is equivalent to 0.43mL/kg. The doses of 0.43mL/kg/bw as well as 0.21 mL/kg/bwadministered to the sub-diabetic and diabetic groups; while 0.43mL/kg/bw was given to stem juice control group. The rats were weighed and the appropriate dose calculated and administered.

Selection of Animal and Animal Care;

35 healthy Albino (Wister strain) rats were acclimatized for a period of 7 days to our laboratory conditions prior to the experiment. Rats were housed in colony cage (5 rats per cage) at room temperature with 12 hours light and 12 hours dark cycle at the Animal House of the Science Laboratory Technology, Federal Polytechnic Idah. Rats had free access to standard foods and water. Handling,

management and use of the animals for the experiment followed the "Ethical and Scientific Considerations Regarding Animal Testing and research.

Induction of Diabetes Mellitus and Feeding of Rats;

Rats were on fasting before the induction of diabetes with alloxan oral feeding. Single oral feeding of freshly prepared solution of alloxan at the dose of 150mg/100mL of normal saline/kg body weight/rat was performed using micro-pipette. This single dose of alloxan produced type II diabetes (having fasting blood sugar level more than 125mg/dL) after 48 hours of alloxan oral feeding and this diabetic state was maintained throughout the experimental schedule. Rats were given standard foods; water and plantain stem juice were administered orally.

Testing of Fasting Blood Glucose Level;

At the time of grouping of the animals, fasting blood glucose (FBG) level was measured. At the interval of two hours, FBG was further recorded from all the animals of all groups throughout the experimental duration. Blood was collected from the tip of tail vein and blood glucose level (BGL) for fasting blood glucose (FBG) and glucose tolerance test (GTT). Studies were assayed using single touch glucometer (Accu-chek Sensor). Depending on their glucose level, the animals were divided randomly into two groups, sub-diabetic rats with FBG of 80mg/dL-124mg/dL and severely diabetic with FBG above 125mg/dL, but both show abnormal glucose tolerance.

Experiments Procedure;

Thirty five rats were divided into seven groups as follows: **Group I** (Normal Control group): Rats of this group received standard foods and water only.

Group II (Stem Juice Control group): Rats of this group received standard foods, water and plantain stem juice were administered orally.

Group III (Diabetic Control group): Rats of this group received standard foods, water and diabetes was induced in rats of this group by single oral feeding of alloxan dose of 150mg/100mL of normal saline/kg body weight/rat.

Group IV (Diabetic Rats): The diabetic rats of this group were forcefully fed with plantain stem juice at the dose of 250mg/kg per body weight/rat/day for 14 days.

Group V (Diabetic Rats): The diabetic rats of this group were forcefully fed with plantain stem juice at the dose of 500mg/kg per body weight/rat/day for 14 days.

Group VI (Sub-diabetic Rats): The sub-diabetic rats of this group were forcefully fed with plantain stem juice at the dose of 250mg/kg per body weight/rat/day for 14 days. Group VII (Sub-diabetic Rats): The sub-diabetic rats of this group were forcefully fed with plantain stem juice at the dose of 500mg/kg per body weight/rat/day for 14 days. Rats of group IV, V, VI and VII received standard foods, water and diabetes was induced in rats of this group by oral feeding of alloxan dose of 150mg/100mL of normal saline/kg body weight/rat. Administration of plantain stem juice in group II, IV, V, VI and VII was performed at early morning and at fasting state. From the starting day of plantain stem juice supplementation to diabetic rats, fasting

blood sugar level of rats in all groups was measured by single touch glucometer at the interval of two hours.

Blood Collection for Biochemical Analysis;

On 16th day of experiment, all the animals were sacrificed by decapitation after recording the final body weight. The rat was deeply anaesthetized. To ensure prolonged anesthesia, a paper towel soaked in volatile anesthetic (Chloroform) was placed on rat's nose during the procedure. The rat was placed on its back, and then a finger was placed at the level of lowest ribs without applying any pressure, the heart is roughly 1 cm above this point, slightly right. Now holding the syringe at 45 degree angle, the needle was inserted between the two ribs and without moving the syringe, the syringe plunger was pulled to fill the syringe with blood. The syringe drew 3 ml of blood from 70-150g rats. The rat was immediately euthanized. The blood samples collected were analysis on the same day of collection and serum was separated by centrifugation at 3000rpm for 5 min for the assay of the blood urea nitrogen and creatinine concentration in the blood samples, using the Cobas 111 instrument.

IV. RESULTS AND DISCUSSION

Results: Semi-automatic Clinical Chemistry Analyzer (Cobas III instrument) was used to determine the serum urea and creatinine concentration in the samples.

Table:1 The results of effect of *Musa parasidiaca* in renal activity in an Alloxan induced diabetic and sub-diabetic rats.

Experiment	Treatment with	Creatinine	BUN
al Rats	Juice	(mg/dL)	(mg/dL)
Stem juice	Feeds +	0.48	5.32
control	500mg/kg/bw/r		
	at/day		
Normal	Feeds only	0.36	5.04
control			
Diabetic	Feeds only	0.53	7.28
control			
Sub-diabetic	Feeds +	0.50	6.72
rats	250mg/kg/bw/r		
	at/day		
Sub-diabetic	Feeds +	0.48	6.44
rats	500mg/kg/bw/r		
	at/day		
Diabetic rats	Feeds +	0.51	6.28
	250mg/kg/bw/r		
	at/day		
Diabetic rats	Feeds +	0.46	5.04
	500mg/kg/bw/r		
	at/day		

Discussion: The table above shows the results of effect of *Musa paracidiaca* in renal activity in alloxan induced subdiabetic and diabetic rats; the observed data shows a slight difference in blood creatinine level between normal control and plantain stem juice control. But when comparing the results obtained from sub-diabetic and diabetic rats both treated with 250mg/kg/bw and 500mg/kg/bw respectively with that of normal control and plantain stem juice control, the results are high. With exception to diabetic rats treated

with 500mg/kg/bw, this increasing effect shows that plantain stem juice could elevate the creatinine level in blood of healthy individuals. When compared with diabetic control, Plantain stem juice shows a lowering effect on the blood creatinine concentration. This decrease blood creatinine level indicates that plantain stem juice could decrease creatinine in the blood of diabetic individuals.

The results on table above also revealed a slight difference in blood urea nitrogen (BUN) between normal control and plantain stem juice control. But when comparing the results obtained from sub-diabetic and diabetic rats both treated with 250mg/kg/bw and 500mg/kg/bw respectively with that of normal control and plantain stem juice control, the results are high. This, in similar case with creatinine test shows a slight increase in BUN level of sub-diabetic and diabetic rats treated with 250mg/kg/bw and 500mg/kg/bw. With exception to diabetic rats treated with 500mg/kg/bw having the same results as that of the normal control. This comparison proves that plantain stem juice is not beneficial for individuals that have normal BUN concentration. The doses of 250mg/kg/bw 500mg/kg/bw produced a significant reduction of BUN level in alloxan induced sub-diabetic and diabetic rats as compared to diabetic control. This shows that there is a good difference in the efficacy of both doses when compared to diabetic control.

V. CONCLUSION AND FUTURE SCOPE

From the results of these analysis, it was found that the blood nitrogen urea (BUN) and blood creatinine concentration lowering effect of plantain stem juice in alloxan induced diabetic and sub-diabetic rats was dose dependent. Much efficacy of plantain stem juice on BUN and blood creatinine level was seen in rats treated with 500mg/kg/bw, while little effect were recorded from test results of rats treated with 250mg/kg/bw. This shows that the higher the dose, the longer the duration of hypomuscular activity which ultimately reduces BUN and blood creatinine concentration.

In conclusion, the effect of the stem Juice of *Musa* paradisiaca suggests that it can be useful in controlling creatinine and BUN concentration in patient suffering from diabetes and will serve as a better alternative to synthetic drugs.

Further analysis should be undertaken to elucidate plantain stem juice metabolism and total excretion from the body and more research should be done to further reveal the efficacy of doses to be administered in the management of diabetic kidney disease.

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