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Hypoglycaemic and Haematologic Effects of Crude Stem Juice of *Costus Afer* on AlloxanInduced Diabetic Wistar Rats

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ABSTRACT

There is a reported surge in the use of different parts of medicinal plants in the treatment of diabetes mellitus in recent times.
Objective: To evaluated the effects of oral administration of crude juice obtained from *Costus afer* stem on the blood glucose concentration and haematological indices of alloxan-induced diabetic wistar rats.
Methods: Forty wistar albino rats weighing between 150g and 250g were used in this study. Twenty rats were treated with 100mg/kg body weight of alloxan by intraperitoneal administration, while 20 rats served as control. The animals were treated with 1.5ml of *Costus afer* crude stem juice.
Results: Results obtained showed that there was a significant ($p < 0.05$) reduction in blood glucose levels of the diabetic animals treated with the crude juice. There was also a slight reduction ($p > 0.05$) in the glucose level of normal animals treated with the juice. Treatment with the juice significantly ($p < 0.05$) altered some haematological parameters of diabetic rats when compared with the control.
Conclusion: Crude stem juice of *Costus afer* possess some protective potential against the effect of diabetes mellitus by reducing the blood glucose level and positively influencing the haematological indices and as such may be employed in the management of diabetes mellitus.
Keywords- Medicinal plants, *Costus afer*, diabetes mellitus, haematological indices.

INTRODUCTION

Diabetes mellitus is a group of metabolic diseases that is characterized by abnormally elevated levels of blood sugar, which perturbs the tightly regulated metabolisms of carbohydrates, proteins and fats arising from defects in the insulin secretion, insulin action or both[1]. Type 1 diabetes mellitus has been attributed to autoimmune destruction of pancreatic β-cells leads to a deficiency of insulin secretion, while type 2 has been attributed to receptor resistance to circulating insulin. Anaemia and other hematologic derangements have been reported to complicated and correlate with

hyperglycemia due to the increased non-enzymatic glycosylation of erythrocytes membrane proteins[2].

Reports have shown that plants are the basis for the development of modern pharmacologic agents and plant parts (roots, stems, leaves, barks, or even fruits have been used for many years to treat diseases, promote and improve health and all over the world[3,1]. Several medicinal plants have been shown from experimental evidences to possess hypoglycemic and antihyperglycemic activities. It has also been reported that about 800 plants could be utilized in the management of diabetes mellitus[1,4]. Moreover, World Health Organization also stated that efforts should be complimented with adequate research in quality, efficacy and safety of medicinal plants.

Costus afer is a pant that belongs to the family *Zingiberaceae*[5]. It is a tall, herbaceous and unbranched tropical plant with creeping rhizome. Omokhua (2011) reported that in Africa, *Costus afer* is found in the forest belt from Sengal to Ethiopia and in the East to Tanzania, Malawi and Angola, in the South and in West Africa[6]. It is common plant in Nigeria, Ghana, Togo, and Cameroun. It is often planted in home garden for medicinal purposes. It is mostly found in moist or shady forest and river banks of West Africa[7,8]. Costus afer is also called bush cane or monkey cane[9]. In Nigeria, plant is known by various local names such as the *Ukhuere-oha* (Bini), *Okpete* or *Okpoto* (Ibo), *tete-egun,* (Yoruba) kakizawa (Hausa) and *mbritem* (Ibibio). The parts of this plants (leaves, stem and rhizome) harvested from the wild plant are commonly used as medicinal herbs in the treatment of various ailments[5,10]. Coctus afer has been reprted to contain bioactive components including diosgenin , saponins aferosides A-C, diosein , parphyllin c, flavonoid, glycoside, kaempterol 3-0¥-Lrhamnopyranoside[6]. The juice from the stem is usually taken orally as antidote for abdominal upsets, cough and respiratory problems[5]. Our decision to carry out this study was informed by the fact that this plant is being used in the management of diabetes mellitus in traditional medicine[11].

MATERIAL AND METHODS

Experimental Animals and plant sample collection

The 40 albino wistar rats weighing between 150g and 250g used in this study were procured from the Animal House of the Faculty of Basic Medical Sciences, University of Uyo, Nigeria. An approval was obtained from the Research Ethical Committee of the Faculty of Basic Medical Sciences of the College of Health, University of Uyo, Nigeria. The animals were kept in cages with stainless steel mesh at the bottom that made the faeces and feed droppings inaccessible to the experimental animals. The animals were fed daily with commercial rat mash and drinking water *ad libitum*. They were fed for two weeks to acclimatize before the experiment.

Matured stem of *Costus afer* were harvested from the forest beside University of Uyo Town Campus. The plant was identified by a Pharmacognosist at the Faculty of Pharmacy, Uninversity of Uyo. The stems were peeled using a knife and then mashed in a motar to bring out the crude juice. The juice was squeezed out from the mashed stem and filtered to obtain a clear juice. The juice was not subject to any form of treatment.

Experimental Induction of Diabetes and Treatment of Animals

Forty albino rats of weight ranging 150-250g were used. Twenty rats were fasted overnight and injected intraperitoneally with 10mg/kg body weight of alloxan monohydrate (Sigma St. Louis, Mo, USA) as 5g/100ml distilled water. The

fact that one of the most potent methods of experimental induction of diabetes is chemical induction by alloxan[12], informed the use of alloxan in this study. After 4 days, 18 surviving rats with fasting blood glucose levels above 14mmol/L were considered diabetic and used for the study[13]. The animals were grouped into treatment groups as follows:

Group 1: Normal control rats received only food and drinking water (NC)
Group 2: Normal treated rats received 1.5ml of fresh crude stem juice of *C. afer* [NT]
Group 3: Diabetic control rats received only food and distilled water [DC]
Group 4: Diabetic rats received 1.5ml of crude stem juice of *C. afer* [DT]

Crude *Costus afer* stem juice was orally administered once day for a period of 14 days. Random and fasting blood glucose levels were monitored every day throughout the administration period using One Touch glucometer. Blood samples were obtained for this by pricking the tip of the tails with a sterile lancet. The prick injuries were cleaned with methylated spirit to avoid infecting the animals. At the end of the treatments, the animals were anesthetized using chloroform and sacrificed. Blood was obtained by cardiac puncture and collected in EDTA containers and numbered accordingly. Analysis of haematological parameters was carried out within one hour of sample collection, with the help of an automated analyzer at the Heamatology Department of the University of Uyo Teaching Hospital.

Statistical Analysis

The results obtained were statistically analyzed using one way ANOVA followed by student t-test. Values were considered statistically significant at $p<0.05$. Data are presented as mean ± standard deviation.

RESULTS

The results of oral administration of the experimental animals with crude stem juice of *Costus afer* are as shown in table1 and table 2 below.

The results of the mean fasting blood glucose levels (mmol/L) of the experimental animals were 4.25±1.86, 4.72±1.84, 26.47±2.54 and 14.35±1.65 for Normal Control (NC), Normal Treated (NT), Diabetic (DC) Control and Diabetic Treated (DT) respectively, while the mean random blood glucose levels (mmol/L) were 12.03±1.86, 10.18±1.86, 33.68±1.86, 20.08±1.82 respectively for NC, NT, DC and DT groups (Table 1). Results of haematological indices (Table 2) showed mean WBC count (x 10/9L) of 10.84±1.40, 13.40±2.93, 8.69±2.00 and 8.40±1.28 for NC, NT, DC and DT groups respectively. Mean RBC counts (x 10/12L) were 6.90±0.72, 7.42±0.53, 7.69±1.16 and 7.33±0.34 respectively for NC, NT, DC and DT groups. The mean PCV (%) were 43.50±3.44, 47.30±1.03, 51.40±1.37 and 34.40±19.70 for NC, NT, DC and DT groups respectively. The mean heamoglobin concentration (g/dl) was found to be 14.10±0.76, 15.30±0.43, 8.30±0.61 and 13.20±0.69 for NC, NT, DC and DT groups respectively. The mean platelet count (x 10/9L) for NC, NT, DC and DT groups respectively, were 750.30±80.00, 803.20 ±60.20, 744.30±130.9 and 749.30±78.10. Other parameters are as shown in Table 2.

Diabetes mellitus is known to be one of the major health challenges all over the globe today[14,15]. It is a disease characterized by hyperglycaemia. Induction of diabetes mellitus in the experimental animals resulted in a significant ($p<0.05$) increase in blood glucose concentration. This may be due to the toxic action of alloxan on the beta cells

of the pancreas via the inhibition of glucokinase enzyme, oxidation of sulphydryl groups, production of free radicals and perturbation of calcium homeostasis[16-18].

In the present study, the effects of the crude stem juice of *Costus afer* on blood glucose kevel and haematological indices of alloxan-induced diabetic rats were investigated. It was observed from this study that experimental induction of diabetes resulted in a significant ($p<0.05$) reduction of white blood cell count. This corroborates with earlier reports by Aladodo *et al.* (2013)[19] and Prophp *et al.* (2008)[20]. However, treatment with this plant extract did not significantly affect the white blood cell count. Experimental induction of diabetes mellitus showed a significant ($p<0.05$) reduction in the hemoglobin and the packed cell volumes of the animals. Treatment with the plant juice significantly ($p > 0.05$) increased the hemoglobin and packed cell volumes of the diabetic animals when compared with the control. Previous research had shown that there are alterations in haematological parameters in the course of diabetes mellitus[21]. Reports also showed that ingestion of medicinal plants can result in the alteration of normal haematological values[22]. It is reported that reactive oxygen species generated during alloxan metabolism is implicated in red cell damage[23]. Also, diabetic rats form glycosylated hemoglobin hence, decreased total hemoglobin [24]. Improvement of the haemoglobin and packed cell volume of the diabetic animals may be due to reduction in oxidative stress on the cell membrane due antioxidants in the plant juice. In this study, induced diabetes did not markedly alter platelet levels and treatment with the plant extract did not show any significant effect on the diabetic animals. However, there was a significant rise in platelets count in the normal treated animals in comparison to the normal control.

Diabetes mellitus is considered to be a prothrombotic state that is characterized by chronic activation of platelets, activation of the coagulation system and decreased fibrinolytic potential[25].

CONCLUSION

It was concluded, from our study, that crude stem juice of *Costus afer* possess some protective potential against the effect of diabetes mellitus by reducing the blood glucose level and impacting positively on the haematological indices and as such may be employed in the management of diabetes mellitus. The mechanisms by which this plant stem juice reduced blood glucose level and alters haematological parameters are subjects for further research.

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Table 1. Blood glucose levels of albino rats treated with crude Stem juice of *Costus afer*

Group	Fasting Blood Sugar (mmol/L)	Random Blood Sugar (mmol/L)
Normal Control (NC)	4.25±1.86	12.03±1.86
Normal Treated (NT)	4.72±1.84	10.18±1.86
Diabetic Control (DC)	26.47±2.54	33.68±1.86
Diabetic Treatment (DT)	14.35±1.65	20.08±1.82

Results are expressed as mean± standard deviation (SD) of 10 determinations.
FBS = Fasting blood sugar,
RBS = Random blood sugar

Table 2. Haematological Parameters of albino rats treated with crude stem juice of *Costus afer*

Parameters	Normal Control (NC)	Normal Treated (NT)	Diabetic Control (DC)	Diabetic Treated (DT)
WBC count (x $10/^{9}$L)	10.84±1.40	13.40±2.93	8.69±2.00	8.40±1.28
RBC count (x $10/^{12}$L)	6.90±0.72	7.42±0.53	7.69±1.16	7.33±0.34
PCV (%)	43.50±3.44	47.30±1.03	51.40±1.37	34.40±19.70
Heamoglobin (g/dl)	14.10±0.76	15.30±0.43	8.30±0.61	13.20±0.69
Platelet count (x $10/^{9}$L)	750.30±80.00	803.20±60.20	744.30±130.9	749.30±78.10
MCV (fl)	63.40±6.01	63.90±4.39	61.70±1.81	62.40±0.63
MCH(Pg)	20.60±1.76	20.60±0.93	21.20±0.41	19.30±0.57
MCHC (g/dl)	32.50±0.86	32.30±1.21	31.80±0.92	31.50±0.82
MPV (fl)	8.23±0.21	8.30±0.89	9.78±0.95	9.85±0.58
PDW	16.00±0.23	15.90±0.48	16.00±0.28	16.10±0.24

Results are expressed as mean± standard deviation (SD) of 10 determinations.
WBC = white blood cell,
RBC = Red blood cell,
PCV = Packed cell volume,
MCV= Mean cell volume,
MCH= Mean cell haemoglobin,
MCHC = Mean cell heamoglobin concentration,
MPV = Mean platelets volume