EFFECTS OF ORAL ADMINISTRATION OF AQUEOUS EXTRACTS OF UNRIPE*Musa paradisiaca*ON ALLOXAN INDUCED DIABETIC ALBINO RATS

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Abstract: The hypoglycemic potential and likely antidiabetic properties of the aqueous extract of unripe *Musa paradisiacal* (plantain) on blood glucose level of wistar rats was investigated and compared with known potent antidiabetic drug (chlorpropamide) in an attempt to encourage exploration of hidden food substances with medicinal properties. Thirty wistar rats were used and divided into six groups of five rats each. Group 1 served as the normal control (positive control) and Groups 2, 3, 4, 5 and 6 were administered with alloxan (100 mg/kg) intraperitoneally. Group 2 served as the diabetic control (negative control). Groups 3, 4 and 5 were orally administered with aqueous extract of *Musa paradisiaca*(140, 180 and 220 mg/kg) once daily for 14 days. Wistar rats in Group 6 were orally administered with chlorpropamide (84 mg/kg) once daily for 14 days. The serum concentration of glucose of all the rats in each group was determined 48 h after inducement of alloxan. This was counted as day one of the test and daily treatment was carried out according to the respective dosages of each group. The serum concentration of glucose of all the rats in each group was again determined after the 7th and 14th dose. There was significant ($p<0.05$) reduction of serum glucose in Groups 3, 4 and 5 that were administered with the aqueous extract of *Musa paradisiaca* after the 7th and 14th dose when compared to the negative control group. Group 6 that was treated with chlorpropamide (84 mg/kg) showed no significant ($p>0.05$) reduction of serum glucose compared to most effective dose of the aqueous extract (220 mg/kg) after the 7th and 14th dose. This result suggests that the aqueous extract of *Musa paradisiaca*possess some hypoglycemic potential and anti-diabetic effect on alloxan induced diabetic rats and thus could be recommended to diabetic patients.

Keywords: Alloxan, antidiabetic, chlopropamide *Musa paradisiaca*, hypoglycemic, wistar rats

Introduction

Diabetes mellitus (DM) is a chronic disorder characterized by impaired metabolism of glucose and lipids due to defect in insulin secretion (beta cell dysfunction) or action (insulin resistance) or both. The characteristic properties of diabetes mellitus are chronic hyperglycemia, microvascular (e.g. retina, renal glomerulus and peripheral nerve) as well as macrovascular (e.g. atherosclerosis, coronary artery disease (CAD), stroke) pathologies with more than 17.5 million deaths worldwide attributable to cardiovascular complications (Banerjee and Vats, 2014). Diabetes is the leading cause of non-communicable diseases worldwide and has reached epidemic proportions in certain parts of the world and in certain ethnic groups (Sudagani and Hitman, 2013). By far, the most common forms of DM are type 1 and type 2 diabetes. Type 1 DM is caused by autoimmune destruction of the insulin-producing β-cells, and type 2 DM is caused by severe insulin resistance and subsequent β-cell failure, due primarily to obesity and lack of physical activity. Other forms of DM include gestational DM and various monogenic types of DM that are caused by single gene defects which lead to deficiencies in β-cell development, insulin production and secretion (Lasselin *et al.*, 2012)

Plantains (*Musa* spp.) are grown extensively throughout the tropical and subtropical regions of the world. The common name is plantain while the botanical name is *Musa paradisciaca*from a family of musaciae and kingdom plantae, genus; Musa and order Zingiberales (Hoffbrand and Moss, 2011). Together they represent the number-one fruit crop in the world, in terms of both production and trade, exceeding oranges by 37 million tons per year for production and by 10 million tons per year for trade. The fruits are slender, angular to pointed, and are generally palatable only after cooking, frying or when ripe (Jekayinfa *et al.*, 2012). The unripe fruit is not accepted by most people as attracted menu particularly the youth and children who prefer sweet foods. However, the adult of a low population eat the unripe plantain cooked with or without pumpkin leaves as porridge and in many other forms. It is also eaten when smoked with palm oil and as chips when fried, which many prefer. The investigation of hypoglycemic potentials of unripe plantain and its likely antidiabetic properties as compared with known potent antidiabetic drug is an attempt to encourage exploration of hidden food substances with medicinal properties. Plantain is rich in fiber, iron, vitamins minerals and serotonin (Jimmy and Okon, 2012).

Materials and Methods

Chemicals and materials

Chemicals used include; alloxan monohydrate, distilled water and chlorpropamide, while the equipment used include; glucometer (Fora V30 model), measuring cylinder, razor blade, weighing balance (Mettler Toledo model MS 204TS), mortar and pestle, sieve and water bath (model PURA)

Experimental design

Animal

Thirty (30) wistar rats were procured from the animal room in the Zoology Department of the Faculty of Science, Bayero University, Kano, Nigeria. The animals used for the study had an average weight between 120 -210 g. The animals were kept in a well-ventilated room in the animal house. They were allowed free access to both food and water throughout the period of study.

Preparation of unripe plantain fruits

Unripe plantains were purchased from Yankura market, Kano, Kano State, Nigeria. Proper arrangement was made concerning its handling and transportation. The plantain was received within 48 h after harvesting.

The unripe plantain was washed, peeled, sliced and air dried. The dried slices were pulverized into powder form and 800 g of it macerated with 3000 ml of distilled water. The mixture was filtered after a day and the filtrate evaporated at 45°C with water bath using methods of Trease (1966).

Preparation of chlorpropamide

Each tablet of chlorpropamide contains 500 mg. Ten (10) tablets of chlorpropamide were dissolved in 50 ml of distilled water following standard solution proparation.

Inducement of diabetes in rats

Alloxan was produced by dissolving 1.2 g in 12 ml of normal saline. The normal saline was prepared by dissolving 0.95 g NaCl in 100 ml of distilled water. Diabetes was induced by single intraperitoneal injection of alloxan monohydrate (100 $mgkg^{-1}$). The animals were allowed 72 h of rest for blood glucose stabilization (Williamson *et al.*, 1996), before the administration of the extract, the initial blood glucose of each of the rats were measured. The volume of the alloxan solution containing 100 mg/kg given to each rat was determined by its weight according to the equation:

$$Volume\ administered(ml) = \frac{weight\ of\ rat\ in\ (\text{kg}) x Dose(mgkg^{-1})}{concentration\ of\ alloxan(mgml^{-1})} \quad \ldots\ldots\ldots\ldots 1$$

Grouping of animals

In the study, thirty (30) animals were used. The animals were divided into six (6), each group consisted of five (5) wistar rats.

Group 1: Had an average weighed of 145 g and served as the positive control and received neither alloxan nor the aqueous extracts of *Musa paradisiaca*.

The rest of the groups received a single dose of alloxan (100 mg/kg).

Group 2: Had an average weighed of 163 g and served as the negative control and received alloxan only.

Group 3: Had an average weighed of 180 g and received 140 mg kg^{-1} body weight of aqueous extracts of *Musa paradisiaca*.

Group 4: Had an average weighed of 187 g and received 180 mg kg^{-1} body weight of aqueous extracts of *Musa paradisiaca*.

Group 5: Had an average weighed of 212 g and received 220 mg kg^{-1} body weight of aqueous extracts of *Musa paradisiaca*.

Group 6: Had an average weighed of 194 g and received 84 mg kg^{-1} body weight of chlorpropamide.

The animals were treated once daily for a period of fourteen days.

Fasting blood glucose determination

Glucometer was used to determine the blood glucose levels of the wistar rats. The glucometer was switched on and the glucose strip inserted in the glucometer and the glucose levels measured in mg/dl after at least each 45 seconds interval.

Collection and preparation of sera samples

The blood of the rats was collected from the tip of the tail, 72 h after being induced with diabetes, and were confirmed to be diabetic if the glucose concentration was ≥180 mg/dl. After seven days of treatment, their blood glucose was determined using a glucometer. Following the last day of treatment i.e. the fourteenth day, the animals were sacrificed and their blood collected in test tubes and the concentration of the sugar was measured using a glucometer.

Results and Discussion

Antidiabetic study

The antidiabetic activity of the aqueous extract of *M. Paradisiaca*was determined in rats administered orally with the extract for 7 and 14 days once daily. The result obtained after 7th dose of treatment with the extract has shown a significantly higher ($p<0.05$) serum level of glucose in diabetic control rats (334.668±18.833) when compared with the normal control rats(100.4±7.162) as shown in Table 1. Of the three groups orally administered with different doses of the extract, Group 3 and Group 4 did not have their serum levels of glucose significantly lower ($p>0.05$) when compared to diabetic control rats, but higher than that of the normal control rats (Table 1). The serum level of glucose in Group 5 had their glucose level lower (232±29.858) when compared to the diabetic control group (334.667±18.33). The serum level of glucose in chlorpropamide treated rats was found to be significantly lower (194±88.042) when compared to the three groups orally administered with different doses of the extract. In the 14th dose of treatment the serum level of glucose was found to be significantly higher in diabetic control rats (285±3.606) when compared to normal control rats (Table 1). The three groups orally administered with different doses of the extract had their serum levels of glucose significantly lower ($p<0.05$) when compared to diabetic control rats, but higher than that of the normal control rats, though there was no significant difference ($p>0.05$) between the normal control rats and Group 5 as shown in Table 1. The serum level of glucose, in chlorpropamide treated rats was found to be significantly lower ($p<0.05$) when compared to Group 3 and Group 4. There was no significant difference in normal control ($p>0.05$) when compared to the Group 5 and the group administered with 84mg/kg of chlorpropamide.

Table 1: Serum glucose level (mg/dl) in alloxan induced diabetic rats before administration of extract, after 7th and 14th dosage of oral administration with aqueous extract of *Musa paradisiaca*and chlorpropamide

Groups	72 Hours After Inducement of Diabetes	7th Dose After Commencement of Treatment	14th Dose After Commencement of Treatment
1. Normal control	100.4±7.162	100.4±10.139	101.8±4.817
2. Diabetic rats without treatment	334.668±18.833*	334.667±18.33*	285±3.606*
3. 140 $mgkg^{-1}$ of extract of *M. paradisiaca*	348.6±50.168*	309.8±16.679*	211.8±15.123* [a]
4.180 $mgkg^{-1}$ of extract of *M. paradisiaca*	383±24.236*	301.4±52.17* [a]	155.2±25.193* [a]
5. 220 $mgkg^{-1}$ of extract of *M. paradisiaca*	356±51.254*	232±29.858* [a]	116.6±14.622*
6. 84 $mgkg^{-1}$ of Chlorpropamide	364.5±97.881*	194±88.042*	95.667±9.292*

Values with asterisk in each column are significantly different at $p<0.05$ compared to normal control; Values bearing superscript (a) in each column are significantly different at $p<0.05$ compared to diabetic control

The significant increase in serum glucose level in diabetic control rats compared to normal control rats is as a result of damage of the pancreatic beta cells by the effect of alloxan. Alloxan monohydrate is one of the chemicals used to induce diabetes mellitus. It induces diabetes by damaging insulin secreting cells of the pancrease leading to hyperglycaemia (Szudelski, 2001). In alloxan induced diabetes, there is selective necrosis of the β-cells of islet of langerhans in the pancreas so that insulin production is totally or partially inhibited, depending on the concentration of the alloxan (Etuk, 2010). The action of reactive oxygen species causes rapid destruction of beta cells (Szudelski, 2001). One of the targets of the reactive oxygen species is DNA of pancreatic islets. Its fragmentation takes place in beta cells exposed to

alloxan (Takasu *et al*., 1991). Following its administration, alloxan is concentrated in the islets and in the liver, where it is reduced to dialuric acid. This acid is unstable in aqueous solutions and undergoes oxidation back to alloxan, accompanied by generation of O^{2-}, hydrogen peroxide and hydroxyl.

Historical records provide a reservoir of basic information on the use of traditional medicine in the management of diabetes mellitus with plant extracts (Srinvasan, 2005). In this study, the comparative studies of antidiabetic potentials of crude extract of *Musa paradisiaca* and chlorpropamide a known diabetic drug has unveiled the high efficacy of both. However, the efficacy of the extract was dose dependent (high dosage). But there was no strong significant difference between Group 3 and Group 4 i.e. the low and middle dosages in terms of the blood fasting glucose. The fasting blood glucose levels decreased as the period of administration of the extract increased. This means that for effective glucose depletion the extract must be taken for a longer period. However, other factors like, the degree of processing e.g., method, time, heat, the starch content (Niba, 2004) may influence the glucose response to the extract (Pi-Sunyer, 2002). The interaction between the glucose and protein may also influence the effect of extract, (Manders *et al*. 2005). The major interest of this study was to determine the antidiabetic potentials of plantain extract and the future approach of likely isolation and purification of the hypoglycemic active ingredient for possible use as an antidiabetic drug. This informed the comparison with chlorpropamide, a potent antidiabetic drug.

Conclusion

The findings of this study indicated that the aqueous extract of *Musa paradisiaca* exert its hypoglycemic potential and antidiabetic effect by lowering blood glucose in alloxan induced diabetic rats. The study has given a lead that medicinal ingredients abound in plants which may be from the leaves, fruits, stems or roots. There's need to explore the potentials of these plants and not just totally depend on orthodox drugs that may not only be as effective as herbal plants but with side effects and complications (Nwafor*et al*., 2005).

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