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ANTIDIABETIC ACTIVITY OF GLADIOLUS PSITTASCINUS IN ALLOXAN INDUCED DIABETIC RATS.

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Abstract

The methanol extract of *Gladiolus psittascinus* bulb was evaluated for its antidiabetic activities in alloxan-induced diabetic rats. Blood glucose levels of the glucose loaded and alloxan-induced diabetic rats were estimated over 180 minutes using the O-toluidine and glucose-oxidase methods. The methanol extract at 1g/kg dose exhibited 16.2% decrease in blood glucose level in the glucose loaded rats and a peak effect of 78.9% in the alloxan-induced diabetic rats. The extract exhibited significant blood glucose lowering effects in the oral glucose tolerance test and type 2 diabetic rats. This study shows a possible beneficial effect of *Gladiolus psittascinus* in the management of non-insulin dependent diabetes (NIDDM).

Keywords: *Gladiolus psittascinus*, hypoglycaemic activity, diabetes mellitus.

Introduction

Diabetes mellitus is a major public health problem. According to WHO reports, more than 176 million patients suffer worldwide and it is estimated that in 2025, there will be about 300 million patients living with this condition. The increase is expected to be 42% in developed countries and 70% in developing countries (WHO, 2006; King et al., 1998). Although different types of hypoglycaemic agents such as thiazolidinediones, insulin, biguanides and sulphunylurea are available; there is growing interest in herbal remedies due to the side effects associated with these therapeutic agents (Prout,1974; Holman and Turner, 1991; Kameswara Rao et al., 1997) beside their limitations in managing the disease effectively (Maggs et al., 1998; Misbin et al., 1997).

Gladilous psittascinus Hook (Iridaceae) an herbaceous plant is commonly known as Maid of the mist' or 'dragon's head lily' and 'Baaka' (Yoruba, South west Nigeria). It occurs virtually throughout the grasslands, savannas and woodlands of sub-Saharan Africa. Ethnomedicinally, it is used as remedy for cold, dysentery, asthma, gonorrhoea and intestinal parasites (Adjanohoun et al., 1991.

In our previous ethnobotanical study, *Gladiolus psittascinus* was used by traditional healers in South West Nigeria as an important recipe for treating diabetes mellitus (Fred-Jaiyesimi, 2007). From this point of view, this study aimed at investigating the antidiabetic effect of *Gladiolus psittascinus* using oral glucose tolerance test and alloxan-induced type 2 diabetic rats.

Materials and Methods

Plant materials

Gladiolus psittascinus bulb (voucher No. FHS 007B) was bought from Falawo Market, Sagamu. Nigeria. Botanical identification was performed at the Department of Pharmacognosy, OlabisiOnabanjo University, Sagamu. The bulbs were sliced, air dried and ground into powder.

Extraction of methanolic plant material

Two hundred grams of the bulb powder were macerated in 80% methanol for four days. The extract was filtered, concentrated to dryness under reduced pressure at a temperature of 40^{o}C. The extract yield was 2.65% w/w (based on the dried starting material).

Animals

Healthy Wister albino rats with average weight of 140g were used for this study. The animals were kept and maintained under standard conditions (12h light and dark cycle and room temperature at 25^{O}C) and were fed with standard pellet diet (Ladokun Feeds, Ibadan) and water *ad libitum*.

Experimetal designs for the oral glucose tolerance test and NIDDM assay are indicated below. The experimental protocols were conducted in accordance with internationally accepted standard guidelines for care and use of laboratory animals.

Experimental design

Animals were divided into four groups (n = 5) and treated orally as follow: group 1, received the plant extract (1g/kg b.w); group 2, received glucose solution (2 g/kg b.w); group 3, received glibenclamide (5mg/kg b.w) and group 4, received water (control group).

Oral glucose tolerance test

Oral glucose tolerance test was performed in overnight fasted (18 hrs) normal rats..Just after glucose administration, the test sample and standard reference drugs were administered. Blood waswithdrawn from the tail of each animal at 0, 30, 60, 90, 120 and 180 mins. The fasting blood glucose levels were estimated by the O-toluidine method (Dubowski, 1962; Frings et al, 1970).

Induction of non-insulin dependent diabetes mellitus (NIDDM).

Non-insulin dependent diabetes mellitus was induced in overnight fasted animals by a single intraperitoneal injection of 60 mg/kg alloxan monohydrate (Sigma Aldrich, UK) solution. Hyperglycaemia was confirmed by the elevated glucose levels in blood and was determined 72 hr after injection.

Hypoglycaemic activity evaluation

The diabetic animals were divided into four groups. Group 1 diabetic rats received *G. psittascinus* extract, Group 2 were administered Glibenclamide (5mg/kg), Group 3 diabetic untreated rats received distilled water (2 ml/kg bw) while Group 4 normal non-diabetic rats also received distilled water (2ml/kg bw). Blood samples were collected via the tail and blood glucose levels was measured at 0, 30, 60, 90, 120 and 180 min with a glucometer (Lifescan, Johnson and Johnson Inc. California).

Statistical analysis

Data were expressed as mean ± SEM. The significance of the differences between the means of the test and control animals was established by the student's t-test.

Result and Discussion

Oral administration of the methanol extract of *G. psittascinus* bulbs at 1g/kg body weight produced significant ($P<0.05$) hypoglycaemic effects in the oral glucose tolerance test at 90, 120 and 180 mins (Table 1). The most pronounced effect was observed at 120 min with 16.2% decrease in blood glucose level. The animals treated with glibenclamide (5mg/kg) showed a significant reduction in blood glucose level after 30 min when compared with the blood glucose level of the untreated glucose loaded rats. In the alloxan–induced diabetic rats, the methanol extract exhibited 78.9% glucose loweringeffect within the first 60 min of administration.

Table 1: Effect of methanol bulb extract of *Gladiolus psittascinus* on oral glucose tolerance test. Values are Mean ± SEM; p < 0.05; n = 5; Figures in parenthesis = % decrease, * Significant difference in glucose level when compared with control glucose untreated values (P<0.05).

Gladiolus psittascinus	MEAN BLOOD GLUCOSE LEVEL (mg/dl).					
Time (minutes)	0	30	60	90	120	180
Group 1 (methanol Extract) 1g/kg. (percentage of reduction)	94.8 ± 1.323	158.2 ± 0.558	151.0 ± 0.294 (3.4 %)	130.2 ± 0.321 (15.5%*)	133.8 ± 0.963 (16.2%*)	131.2 ± 2.094 (14.2%*)
Group 2 Glibenclamide (5mg/kg). (percentage of reduction)	81.5 ± 0.006	132.5 ± 0.014 (12.3)	98.9 ± 0.006 (36.7%*)	92.4 ± 0.008 (40%*)	99.8 ± 0.003 (37.5%*)	88.1 ± 0.005 (42.4%*)
Group 3 Glucose untreated Control.	89.1 ± 0.057	151.3 ± 0.031	156.3 ± 0.074	154.0 ± 0.037	159.7 ± 0.093	152.9 ± 0.375
Group 4 Normal (non-diabetic).	85.3 ± 0.085	80.1 ± 0 .437	82.1 ± 0.050	84.8 ± 0.150	85.0 ± 0.044	85.3 ± 0.075

Table 2: Effect of methanol bulb extract of *Gladiolus psittacinus* on blood glucose level of alloxan- induced diabetic rats.

***Gladiolus psittacinus* extract.**	Blood glucose level (mg/dl).					
Time (minutes).	0	30	60	90	120	180
Group 1 extract (1g/kg)	145.0 ±6.816	123.0 ± 2.225 (61.3 %)*	124.6 ± 1.472 (78.9 %)*	119.0 ± 1.608 (69.6 %)*	119.3 ±4.816 (70.2 %)*	116.3 ±0.117 (75.6 %)*
Group 2 Glibenclamide (5mg/kg)	220.1 ±0.254	187.6 ± 0.112 (61.7%)*	175.4 ±0.005 (60.4%)	131.5 ±0.071 (69.5%)*	126.9 ±0.319 (69.7%)*	120.5±0.215 (70.2%)*
Group 3 Diabetic untreated (Control).	300.0± 3.360	489.3± 0.992	443.3 ± 1.216	431.0± 0.627	418.2 ± 0.008	404.7± 0.581
Group 4 Normal (non-diabetic)	88.0 ± 0.246	91.3± 0.275	92.2 ±0.134	87.0 ± 0.103	89.1 ± 0.531	90.1± 0.240

Values are Mean ± SEM; P<0.05; n = 5; Figures in parenthesis = % decrease,* Significant difference in glucose level when compared with control diabetic untreated values (P<0.05).

The standard reference drug (glibenclamide) at a dose of 5mg/kg exhibited 61.7% decrease and a peak effect of 70.2% at 180 min. The effect of the methanol extract and glibenclamide in the diabetic rats when compared with the control (untreated diabetic rats) showed similarity in the activity of the two treatments. The data of the present study shows that *G. psittascinus* may have beneficial effect in non-insulin dependent diabetes mellitus. Methanol extracts are known to contain chemical compounds each of which is capable of producing definite biological activities (Ojewole, 2003), the methanol extract of *G. psittascinus* exhibited a pronounced glucose lowering effect in the alloxan-induced diabetic rats. Although the exact mechanism of action of the extract is unknown, the effect exhibited suggests a possible stimulation of insulin release from the residual β-cells and glucagon inhibition. In addition, the extract might have an insulin-like effect acting by improving the glucose uptake and metabolism or by inhibiting gluconeogenesis thereby exerting the hypoglycaemic effect. Further studies needs however to be carried out to identify the exact mechanism of action of *G. psittascinus* and the compound(s) responsible for the hypoglycaemic activity.

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