The effect of aqueous root extract of *Sphenocentrum jollyanum* on blood glucose level of rabbits

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The anti-hyperglycaemic effect of the aqueous root extract of *Sphenocentrum jollyanum* (SJ) Pierre (Menispermaceae) was evaluated in normal and alloxan-induced diabetic rabbits. The levels of glucose were determined at different doses and times following treatment with the extract. The oral single dose administration of the extract in OGTT study led to significant (p < 0.05) decrease in peak values and the area under curve by 10.5% compared to the untreated. Although the glucose level showed gradual decline it nonetheless failed to return to baseline glycaemia (65.7±4.1) with a value of 118.4±3.9 after 4 h indicating less glucose tolerance as compared to glibenclamide in which 65.7±2.0 was recorded. In alloxan diabetic rabbits SJ exhibited dose dependent significant (p < 0.05) reduction in blood glucose level from day 3 of daily treatment with the extract. The maximum decrease from 337.4±8.9 to 269.6±3.8 was observed in the group that received 200mg/kg⁻¹. This suggests that the plant may be a potential source for the development of a new oral anti-diabetic agent.

Key word: Ant-diabetes, *Sphenocentrum jollyanum*, rabbits.

INTRODUCTION

Diabetes mellitus (DM) is a heterogeneous disorder that is characterized by the derangement of carbohydrate, protein and lipid metabolism. The world has witnessed increase in the incidence of DM in which about 190 million people representing over 3% of the population have been diagnosed of the disease with the figure projected to exceed over 300 million by the year 2025 (Ravid and Rachmani, 2005).

DM is known to comprise of two major types; type 1 and 2, each with distinct pathogeneses (Skyler, 2004). Common to both, however, is hyperglycaemia and various life threatening complications (Abraira et al., 1995; Ohkubo et al., 1995). Although significant success have been recorded with the introduction of insulin (Goldfine and Youngren, 1998) and oral hypoglycaemic agents like sulphonylureas and biguanides (Al-Awaidi et al., 1985; Mariam et al., 1996), currently available drugs particularly for type 2 diabetes have a number of limitations such as significant side effects and high rate of secondary failure (Xie et al., 2005). Consequently, the disease and its related complications are increasing unabated (Tiwari and Madhusudana, 2002).

This means that DM is likely to remain a significant threat to public health in the years to come unless more aggressive intervention is sought. With intensified search for more effective, cheap and readily available therapy, herbal medicine has proved to be useful compliment. For various reasons, in recent years, the popularity of complimentary medicine in the area of metabolic disorder has increased. This is based in part on the available scientific data on medicinal plants including those with anti-diabetic potential (Grover et al., 2002).

In traditional medicine, DM is treated with diet, physical exercise and medicinal plants. Plant extracts are frequently considered to be less toxic and freer from side effects than synthetic drugs (Bailey and Day, 1989). Over the last 10 years, the use of herbs has tripled (AndradeCetto and Heinrich, 2005) owing to the success recorded. As more research interests are tailored towards
plant sources, a recent data base showed that over 389 medicinal plants are currently in use for diabetes mellitus management (Padvala et al., 2006). Different parts of plants have exhibited hypoglycaemic activity from normoglycaemia and also anti-hyperglycaemic effect in response to increase in blood sugar level. The active components of these plants are usually isolated for direct use as drugs, lead compounds and pharmacological tools (Grover et al., 2002).

The impact is phenomenal as they have proved more effective than synthetic drugs in the management of diabetes complications (Li et al., 2004). *Sphenocentrum jollyanum*, a perennial plant is an erect shrub that belongs to the family Menispermacae (Burkill, 1985; Nia et al., 2004). It is distributed along the west coast of Africa from Sierra Leone across Nigeria to Cameroon (Nia et al., 2004). The plant is traditionally used as remedy for feverish conditions, cough and wound dressing and as an aphrodisiac (Dalziel, 1937; Irvine, 1961). Studies have shown the leaves to possess significant antipyretic and analgesic activities (Muko et al., 1998). The roots and leaves have been reported to be active against Polio Type-2 virus (Moody et al., 2002). Investigations revealed that different parts of the plant exhibited significant antioxidant (Nia et al., 2004) and anti-inflammatory (Moody et al., 2006) properties.

Diabetes mellitus is characterized by the alteration in endogenous free radicals scavenging defenses that leads to ineffective scavenging of reactive oxygen species resulting to oxidative damage and tissue injury (Oberley, 1988). The pancreas, with inherent lack of anti-oxidant protection, is usually prone to oxidative stress (Lenzen et al., 1996; Tiedge et al., 1997) that may initiate a sequence of events leading to diabetes mellitus. *S. jollyanum*, having demonstrated capacity to mop up free radicals as well as set up anti-oxidant defenses, stimulated our interest for a pilot investigation of its anti-diabetic potential.

**MATERIALS AND METHODS**

**Plant material**

Fresh roots of *S. jollyanum* were collected from a farm land located in Ijebu-Orou community, Ogun State, Nigeria. The collection was in the month of November, washed with tap water before dried under the sunlight for seven days and further dried in an oven regulated at 40°C. The authentication of the plant was by a taxonomist, Dr. O. A. Ugboogu, Chief Research Officer at the Forest Research Institute of Nigeria (FRIN) where voucher specimen of the plant has been deposited in the herbarium (FHI/108203).

**Preparation of the plant aqueous extract**

The fresh roots were chopped into pieces and air dried (36-39°C). The dried root was ground to a coarse powder with grinder. The powder (2370 g) was placed in a Soxhlet extractor in batches and extracted with water in three cycles for about 60 h. The extracted material was filtered with Whatman filter paper No. 4. The filtrate obtained was dried *in vacuo* between 30-36°C. The yield about 54 g was stored at 4°C under refrigerated condition till needed for.

**Animals**

Healthy adult rabbits of either sex weighing between 1.5-1.8 kg were obtained from the Animal House of the University of Ibadan, Oyo State, Nigeria. Having certified their health conditions, were kept in aluminum cages under standard conditions at room temperature (21±2.8°C) with 12 h dark and 12 h light circle in the Laboratory Animal Centre of the College of Medicine, University of Lagos, Lagos, Nigeria. They were fed standard rabbit pellets from Livestock Feeds PLC, Lagos and water *ad libitum*. The ethical use of the animals was sought and obtained from the animal department of College of Medicine, University of Lagos, Nigeria.

**Induction of diabetes**

Rabbits fasted overnight (18 h) were induced with a single intravenous injection of 150 mg/kg⁻¹ alloxan monohydrate dissolved in sterile water through the auricular vein (Khosla et al., 1995) with modification. Hyperglycaemia was confirmed where elevated blood glucose level was ≥ 250 mg/dl after 72 h of injection (Olajide et al., 1999).

**Effect of extract on alloxan diabetic rabbits**

The diabetic animals were randomized to the following groups of 5 rabbits each: group I was diabetic control; group II received glibenclamide (10 mg/kg⁻¹) orally; groups III, IV and V received 50, 100 and 200 mg/kg⁻¹ of the extract respectively orally that was prepared using 2% tween 80 solution dissolved in distilled water. Treatment was continued for 15 days. Before the treatment (day 0) and after the treatments (days 3, 5, 7, 9, 11, 13 and 15) blood was collected from the rabbit auricular vein, between 0.6 to 0.8 ml and centrifuged to obtain blood plasma which was used for glucose estimation by oxidase method (Olajide et al., 1999).

**Effect of the aqueous extract on oral glucose tolerance (OGTT)**

The rabbits were fasted for 18 h and were randomized to 3 groups of 5 rabbits each. Blood was collected pre-treatment from each rabbit to determine fasting blood glucose. The rabbits in group 1 received 2 ml/kg⁻¹ of distilled water orally. Group 2 received 1 g/kg⁻¹ of the aqueous extract while group 3 received 0.01 g/kg⁻¹ of glibenclamide by gavage. Rabbits in the three groups were given oral glucose load of 1 g/kg⁻¹ (Perfumi et al., 1991) 30 min after distilled water, aqueous extract or glibenclamide administration. Blood was collected from the animals at 0.5, 1, 2, 3 and 4 h after the oral glucose load for the blood glucose estimation (Moshi et al., 1997).

**Phytochemical analysis**

Phytochemical screening was carried out on the extract using the method of Trease and Evans (1989) to identify the possible active ingredients present in the root of the plant.

**Acute toxicity**

Rabbits were divided into five groups of 5 animals each. Different doses (500, 1000, 2000, 4000 and 8000 mg/kg⁻¹) of the aqueous extract was administered by gavages. The animals were observed...
RESULTS

Activities in hyperglycaemic rabbits

Glucose tolerance was evaluated by OGTT. As shown in Figure 1, following oral glucose load in the untreated group (vehicle group), hyperglycaemia occurred which reached the peak level 1 h after the administration. Although the glucose level showed gradual decline thereafter, it nonetheless failed to return to baseline glycaemia after 4 h indicating glucose intolerance. The root aqueous extract of SJ and glibenclamide significantly (p < 0.05) reduced the peak values and the area under curve by 10.5 and 43.9% respectively compared to the untreated. However, glibenclamide exhibited more effective glucose tolerance which caused a return to baseline glycaemia (65.7±2.0) 4 h later unlike the extract treatment in which it was significantly higher (118.4±3.9).

Activities in alloxan-hyperglycaemic rabbits

Table 1 shows the effect of various treatments on blood glucose levels. The root aqueous extract of SJ exhibited dose dependent, significant (p < 0.05) reduction in blood glucose level from day 3 of daily treatment in alloxan induced diabetic rabbits. The maximum decrease occurred at day 15 with values of 291.5 ± 6.1, 280.4 ± 4.4 and 269.6 ± 3.8 (50, 100 and 200 mg/kg⁻¹) respectively while more marked decrease (244.7 ± 2.7) was observed with glibenclamide treatment. The result therefore shows that the extract treatment demonstrated considerably less anti-hyperglycaemic activity compared to the control drug.

Phytochemical analysis

The active compounds found in the extract include; alkaloid, saponins, terpenoid compound, anthraquinones, flavonoids, tannins, cardiac glycosides and reducing sugar.

Acute toxicity test

The extract administered orally up to the highest dose tested (8000 mg/kg⁻¹) produced no mortality. The animals did not manifest any sign of respiratory distress, restless-
Table 1. Plasma glucose level of rabbits treated with SJ root aqueous extract.

<table>
<thead>
<tr>
<th>Dose (mg/kg)</th>
<th>Plasma glucose levels (mg/100ml) during treatment with the extract</th>
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<tbody>
<tr>
<td></td>
<td>Day 0</td>
</tr>
<tr>
<td>Non-diabetic</td>
<td>78.2±7.8</td>
</tr>
<tr>
<td>Diabetic untreated</td>
<td>334.0±8.5</td>
</tr>
<tr>
<td>Glibenclamide 10</td>
<td>350.5±8.8</td>
</tr>
<tr>
<td>S. jollyanum root 50</td>
<td>335.4±10.3</td>
</tr>
<tr>
<td>S. jollyanum root 100</td>
<td>331.9±7.8</td>
</tr>
<tr>
<td>S. jollyanum root 200</td>
<td>337.4±8.9</td>
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</table>

Table shows the plasma glucose concentration during 15 days of extract/glibenclamide administration or 10 ml/kg distilled water (control). Values are Mean ± SEM; n=5, *p<0.05 compared to control (Student’s t-test).

DISCUSSION

In this study, aqueous root extract of *S. jollyanum* exerted significant anti-hyperglycaemic activity with the reduction of the peak glucose concentration in OGTT curve by 10.5% as compared to the untreated. The precise mode of action is yet to be determined. However, it is not unlikely that the plant extract may have stimulated the beta cells to release more insulin. In this light, a number of other plants have been reported to have anti-hyperglycaemic and insulin release-stimulating effect (Venkateswaran and Pari, 2002; Latha and Pari, 2003, 2004; Park et al., 2008).

In alloxan-induced diabetic animals, significant decrease in glycaemia was observed from day 3 of oral administration of the extract to the last day of treatment. However with the maximum decrease of 269.6 ± 3.8, it was obvious that the blood glucose failed to return to basal glycaemia.

Alloxan is known to be specifically cytotoxic to pancreatic beta cells resulting to their death and decrease in endogenous insulin release (Omamoto et al., 1981). It was not unlikely that the beta cells destruction affected the activity of the extract. This was probably also indicative of the extract less effectiveness in lowering the plasma glucose level to basal glyceamia. However the glycaemic decrease that occurred may have been due to the stimulatory effect of the extract on the surviving beta cells to promote insulin release. The extract may have acted in a mode similar to glibenclamide which has been reported to be effective in moderate streptozotocin-induced diabetic animals and ineffective in severe diabetic rats (Ivorra et al., 1988; Sharma et al., 1997). The antidiabetic activity of the extract could be due to the influence of certain active principles like saponins known to be bioactive against diabetes (Abdel-hassan et al., 2000). Furthermore, flavonoids, glycosides and alkaloids frequently implicated with this activity (Loew and Kaszkin, 2002) were also components of the plant extract.

**Conclusion**

The result of this finding clearly indicated that the aqueous extract of *S. jollyanum* root was effective in reducing the blood glucose concentration of alloxan diabetic and hyperglycaemic rabbits. This is informative requiring that further work be conducted. This may be a potential source for the development of a new effective oral anti-diabetic agent.

REFERENCES


