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Short Communication

Exploration of the anti-diabetic effect of the matured seeds of Dioclea reflexa

Adepoju Adeyinka GK* and Balogun O.M.

Department of Clinical Pharmacy and Biopharmacy, Faculty of Pharmacy, Olabisi Onabanjo University, Sagamu, Ogun State, Nigeria.

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The study aimed at exploring the anti-diabetic effect of the seed flour of Dioclea reflexa on alloxaninduced diabetic rats. Thirty Wistar rats of both sexes were exposed to respective doses of the seed flour and the standardized drugs (Gliclazide 6.1 mg/kd body weight and Metformin 60 mg/kg body weight) according to the group specifications. The seed flour at a dose of 2 g/kg body weight per oral once daily was found to be more effective than any of the standard drugs with which it was compared. The result showed that D. reflexa seed flour possessed potent oral hypoglycaemic property probably mediated via increased peripheral utilization of glucose.

Key words: Dioclea reflexa, anti-diabetic, anti-hyperglycaemia, seed flour, hypoglycaemic, glucose utilization, alloxan-induced diabetes.

INTRODUCTION

Despite the great strides made in the pathophysiology and management of diabetes mellitus, the graph of diabetes-related mortality is rising unabated. Although a number of synthetic drugs are available for therapy, the disease and its related complications still remain uncontrolled (Dewanje et al., 2008).

Traditional medicinal remedies have been used since ancient period to treat the ailment thus presenting a stirring prospect for expansion of an alternative way of treating diabetes and its complications (Bailey and Day, 1989). Hitherto, herbal drugs are also prescribed even when their biologically active compounds are unknown (Rahman and Zaman, 1989; Valiathan 1998). Diabetes mellitus represents one of the major diseases associated with increased risk of heart disease, stroke, kidney disease, retinopathy, neuropathy, ulceration and gangrene of extremities (Momin, 1987). Recent statistics showed that the global epidemic of diabetes mellitus is worse and greater in developing than the developed countries (Rotshteyn and Zito, 2004; Oputa 2002). The WHO Expert Committee on diabetes mellitus has recommended the evaluation of the folkloric methods of managing the disease (Oputa, 2002; Adeneye and Agbaje, 2008). It is in the pursuit of this challenge several medicinal plants are being investigated for their hypoglycaemic effects.

It is in an attempt to increase the available traditional remedies and possible further studies to identify the active principles that prompted this study. Hence, the choice of Dioclea reflexa for antidiabetic screening and testing in rats. Further chemical studies on the plant may

*Corresponding author. E-mail:monarch012002@yahoo.com.Tel: 0803-474-0000.

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Groups/drugs	0 h	24 h	48 h	72 h	
1 Normal saline	125.2±1.9	129.4±2.8	124±1.52	107.2±8.5	
2.Diabetic control	214.2±11.7	212.8±8.1	182±19.9	191.6±15.3	
3. Gliclazide (6.1 mg/kg boy weight)	187.2±4.22	89.6±2.11	82.2±0.9	37.8±4.9	
4.Metformin (60 mg/ kg body weight)	206.4±10.5	189±29.15	113.6±12.8	103.6±3.1	
5.Seed Flour(1 g/kg body weight)	190± 2.5	95.7±2.6	85.7±7.5	45.2±6.2	
6.Seed Flour(2 g/kg body weight)	172.4±8.7	65.2±4.33	58.4±6.1	58.4±7.5	

Table 1. Shows the effect of various doses on blood glucose over a72-hr period on the rats.

increase and enhance therapy. The seeds of *D. reflexa* are found in areas of Africa, Asia and some other parts of the world. It is called ARIN (Yoruba) in Nigeria. The antimicrobial effect of the leaves had been examined (Adeneye et al., 2006). There is available evidence of its nutrient composition (Ogundare and Olorunfemi, 2007; Akinyede et al., 2006; Aiyesanmi and Oguntokun, 1996). There is no available evidence of its use in the treatment of Diabetes mellitus but it has been found to have a high fibre content, which makes it a possibility for its incorporation into the diets of diabetics.

MATERIALS AND METHODS

Plant material

Samples of the seeds of *D. reflexa* (Fabaceae) were identified and validated at the Taxonomy section of the Forestry Research Institute of Nigeria (FRIN), Ibadan, Nigeria. The specimen was issued a Voucher Number FH1108765. The seeds were shadedried at room temperature before cracking. Cracking was to remove the pericarp and the seed coat. The cracked seeds were further shade-dried for ten (10) days before crushing and milling in a mechanical grinder to fine powder of mesh 40. The phytochemical screening confirmed that of Akinyede et al. (2006) as containing tannins, proteins, flavonoids, terpenoids and sugars.

Animals

Thirty Wistar rats of both sexes weighing 85 to 125 g were purchased from the Animal House, Pre-clinical Department, College of Medicine, University of Ibadan, Nigeria. They were fed with adequate ration of standard rat feed and water for 30 days under standard laboratory conditions. The rats were randomly assigned into six groups of equal membership. The rats have not participated in any experimental study before.

Experimental induction of diabetes in rats

Diabetes was induced by intra--peritoneal administration of alloxan at the dose of 150 mg/kg body weight to the rats except the Group 1 (Normal saline control). The animals were fasted for 24 h before the administration of the respective group doses as follows:

Group 1 (Normal saline group): Rats were treated with normal saline by injection.

Group 2 (diabetic control): Rats were treated with alloxan 150 mg/kg body weight via tail injection.

Group 3: Rats were treated with Alloxan 150 mg/kg body weight via tail vein injection and after 2 days rats were treated with Gliclazide 6.1 mg/kg body weight daily orally for 3 days.

Group 4: Rats were treated with Alloxan 150 mg/kg body weight via tail vein injection and after two days, rats were treated with Metformin 60 mg/kg body weight daily orally for 3 days.

Group 5: Rats were treated with Alloxan 150 mg/kg body weight and after 2 days, rats were given 1 g/kg body weight Dioclea reflexa daily orally for 3 days.

Group 6: Rats were treated with Alloxan 150 mg/kg body weight via tail vein injection and after two days, rats were treated with Dioclea reflexa 2 g/kg body weight daily orally for 3 days.

Blood glucose determination

The blood glucose levels for the groups were determined at 0, 24, 48 and 72 h intervals using a Liberty Glucometer and test strips. Hourly percentage changes in blood glucose levels were determined for the Group 6 animals to check the rate of change of the glucose levels. Blood was collected from the tail vein periodically as stated.

Statistical analysis

The data obtained from the study were expressed as mean \pm standard error of the mean (SEM). A one-way analysis of variance (1-way ANOVA) was used for the analysis. The statistical level of significance of the difference between the mean of the control and the treated groups was at p<0.05.

RESULTS AND DISCUSSION

Table 1 shows the results of the administration of the seed flour of *D. reflexa* on Wistar rats. In this study, the hypoglycaemic activity of *D. reflexa* (Fabaceae) seed flour was compared with the relative standard activity levels of Metformin and Gliclazide in animal model (alloxan-induced diabetic rats) over a 72 h period. Aloxan induces diabetes by destroying the beta cell of the islet of Langerhans and impairing renal functions (Yusuf and Lasisi, 2006).

From Table 1, it was found out that the seed flour of *D. reflexa* at a dose of 2 kg/kg lowered the blood glucose better than any of the standard anti-diabetic drugs with which it was compared. It also lowered the blood glucose levels when compared to the controls (Groups 1 and 2). These reductions were statistically significant when compared to the other groups (p<0.05). At 1 g/kg body

Table 2. Showing the percentage change in blood glucose levels over a 72 h interval.

Hours	0	1	3	6	24	48	72
Mean±SEM	172.4±8.7	102.6±4.2	116.2±0.58	87.8±12.0	65.2±4.33	58.4±6.1	58.4±7.5
% change in blood glucose		-59.5	-32.6	-50.9	-62.2	-66.12	-66.12

weight of the dose of the seed flour, it was not better than Gliclazide (Group 3) in reducing blood sugar levels. However, it reduced the blood sugar level better than Metformin (Group 4) and the other groups (Groups1 to 3) and this was statistically significant (p<0.05).

It is of note that all the standard drugs and the seed flour showed a statistically significant reduction of blood glucose levels when compared to the control groups (Groups 1 and 2). Table 2 showed the percentage change in the reduction of blood glucose levels in the alloxan-induced hyperglycemic rats when administered with 2 g/kg body weight of the seed flour of *D. reflexa*.

The result indicated a progressive reduction in the blood glucose levels. The percentage range of 32.6% to 66.12% showed the high activity of the seed in reducing blood glucose levels in hyperglycemic rats.

The rapid reduction of the glucose levels under 24 h for both 1 and 2 g/kg body weight doses showed the high activity and ability of the seed flour as a potential antihyperglycaemic agent. This observed hypoglycaemic effect of the *D. reflexa* seed flour was an indication that it contained active principles with potent hypoglycaemic property.

All the animals survived throughout the experiment. Hence, no toxicity-related mortality was recorded throughout the experiment. This seed flour could be considered safe on acute oral administration. Thus, Bruce (1985, 1987) and American Society for Testing and Materials (1987) showed that any chemical substance with LD $_{50}$ estimate greater than 2000 to 5000 mg/kg/oral route could be considered safe and of low toxicity. Thus, lack of toxicity or associated lethality (mortality) with the administered high dose of the seed was an indication of the safety of the seed flour. Further studies need to be carried out histologically to test the toxic effects of the consumption of the seeds of *D. reflexa* on the vital organs.

Conclusion

From the results of this study, it could be inferred that the seeds of *D. reflexa* possessed potent oral hypoglycaemic property probably mediated via increased peripheral utilization of glucose (Yusuf and Lasisi, 2006; Subramonian et al., 1996). Phytochemical screening and isolation of the active principles contained in the seed need to be carried out to know the exact mode of action of the seeds as an oral hypoglycaemic agent.

Conflict of interest

The authors report no declarations of interest.

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