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Antihyperglycemic activity of the ethanolic extract of *Curculigo orchioides* Gaertn

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ABSTRACT

The investigation was carried out to study effects of *Curculigo orchioides* Gaertn rhizome on blood glucose level. The antihyperglycemic efficacy of ethanolic extract of the rhizome was evaluated in normal, glucose loaded and alloxan induced diabetic rats. The extract exhibited significant hypoglycemic activity in all the three animal models when compared with the control group. The activity was also comparable to that of the effect produced by a standard antidiabetic agent Glimperide, 500 µg/kg (p.o). The results also indicated dose dependent effect. The hypoglycaemia and antihyperglycaemia produced by the extract may be due to increased uptake of glucose at tissue level or increase in pancreatic β-cell function or due to inhibition of intestinal absorption of glucose. The study indicated that ethanolic extract is a potential antidiabetic agent and lends scientific support for its use in folk medicine.

KEYWORDS - *Curculigo orchioides*, Glucose tolerance, Glimperide, Antidiabetic, Alloxan

INTRODUCTION

Diabetes mellitus (DM) is a group of metabolic diseases characterized by hyperglycemia, hypertriglyceridaemia and hypercholesterolemia, resulting from defects in insulin secretion action or both (1, 2). Regions with greatest potential are Asia and Africa, where diabetes mellitus (DM) rates could rise to 2-3-folds than the present rates. Many herbal medicines have been recommended for the treatment of diabetes. Plant drugs are frequently considered to be less toxic and free from side effects than synthetic ones (3).

Curculigo orchioides Gaertn Family Hypoxidaceae (Amaryllidaceae) is well known plant in traditional Indian system of medicine Ayurveda. Found in India in the sub-tropical Himalayas from Kumaon eastwards and in the Western Ghats from Konkan southwards. Its tuberous roots are slightly bitter and mucilaginous and considered tonic, alternative demulcent, diuretics and restorative (4). The plant finds mention in Ayurvedic treatise like 'Sushruta Samhita' and 'Charak Samhita' as Rasayan or rejuvenator (5). The drug is also useful as a medical cure for piles, asthma, jaundice, diarrhoea, colic and gonorrhoea (6). The plant has been proven as hepatoprotective (7, 8) immunostimulant (9, 10) and antioxidant (11). Indigenous medical practitioners of Korwa and Khasi hills in M.P. use Kali mustli (*C. orchioides*) in asthma,

jaundice and male sterility (12). Tuberous roots are used as tonic for strength, vigour, vitality and find use in several restorative and aphrodisiac formulations in the Ayurvedic system of medicine (13).

C. orchioides chiefly contains carbohydrates (14), alkaloids, glycoside, saponins and sterols (15). Curculignin B-C, Curculigosaponin-F has also been isolated from *C. orchioides*. The medicinal property of the herb has been mainly attributed to curculigosides and curculigosaponins (16). In Unani system it is used for treating diabetes.(17)

Although acclaimed traditionally as antidiabetic there are no scientific studies regarding the effect of *C. orchioides* on blood glucose level. In the present investigation, the antidiabetic activity of ethanolic extract of *C. orchioides* rhizome is being reported

MATERIALS AND METHODS

Animals

Albino rats (druckery strain) of either sex weighing 120-150 g were fed on standard diet and water *ad libitum*. The animals were housed at room temperature (24± 2 °C) on a reversed day-night cycle (06:00 hrs to 18:00 hrs.). The animals described as fasted were derived of food for 16 h.

Plant Material

Rhizomes of *Curculigo orchioides* Gaertn were collected at Sagar M.P. (India) and taxonomically identified at the Department of Botany, Dr H. S. Gour

University, Sagar. A voucher specimen of the same has been made and deposited (No. NSC-CO-2005) at departmental herbarium.

Chemical preparation

Glimeperide was obtained as gift sample from Sun Pharma, Mumbai. 0.005 % suspension of Glimeperide was also made with 0.2% acacia. The glucose oxidase kit used was that of Techo Diagnostic Placentia. Gum acacia was purchased from CDH. All the animal experimentations were carried after prior permission from the institutional ethical committee of the Dr. H.S. Gour University, Sagar (M.P.) India.

Preparation of extract

The powdered dried rhizomes were defatted by extraction with petroleum ether (60-80 °C). The defatted plant material was then extracted with ethanol (95%), and dried under vacuum (4.08 % w/w). A suspension of ethanolic extract was prepared using 0.2% gum acacia to give 1 % suspension. Ethanolic extract up to 10 gm/kg is as nontoxic (9). We used 100 mg /kg and 200 mg/kg for evaluating the antidiabetic activity.

Method

Effect of alcoholic extracts in normoglycaemics and glucose tolerance in rats.

Fasted rats were divided into five groups of six rats each. Group I serves as normal control. Group II (Extract treated) received 100mg/kg (p.o) extract of plant. Group III (glucose control) received distilled water. (p.o). Group IV and Group V received 100mg/kg (p.o) extract of plant and Glimeperide 500 µg/kg (p.o). After 30 min of extract and standard drug administration, the rats of all groups (III, IV, V) were orally treated with 2g/kg of glucose.

Blood samples were collected from retro orbital plexus at 30 and 90 min after glucose loading. Serum was separated by centrifuge the blood at 2000 rpm for 15 minutes and serum glucose level was determined by Glucose oxidase Method (Kit Techo Diagnostic) (Table 1).

Effect of *C. orchioides* extracts on alloxan-induced hyperglycemia.

Hyperglycemia was induced by a single i.p injection of 120 mg/kg of alloxan monohydrate in sterile saline. After 5 days of alloxan injection, the diabetic rats (glucose level >350 mg/dl) were separated (18) and divided into four groups of six animals each. Group I served as diabetic control and was given distilled water. Groups II-III were treated orally with ethanolic extracts at a dose of 100mg/kg and 200 mg/kg, and group IV received standard drug Glimeperide 500 µg/kg

(p.o). Blood samples were collected from retro orbital plexus at zero time (before receiving the extract, Glimeperide) and 1 and 3 h following the treatment. Serum was separated by centrifuge the blood at 2000 rpm for 15 minutes and serum glucose level was determined by Glucose oxidase Method (Kit Techo Diagnostic) (Table 2).

Statistical analysis

Results are expressed as Mean ± S.E.M. The significance of the data was evaluated using Student's t- test and were considered significant statistically significant when P<0.05. The statistical analysis was carried out using InStat 2.1 software.

RESULTS AND DISCUSSION

In the present study, the hypoglycemic activity of crude extract from *Curculigo orchioides* rhizomes was evaluated in normal, normal hyperglycemic and alloxan-induced diabetic rats. The effect of *Curculigo orchioides* extract on glucose tolerance and normal rat are summarized in table 1, the treatment of normal rats with ethanolic extract and Glimeperide, a known hypoglycemic drug, resulted from a significant decrease ($P < 0.001$) in blood glucose levels 90 min after oral drug administration when compared with initial level and control. The maximum Glucose tolerance was observed at the 30th min for ethanolic extract.

The blood glucose levels of diabetic rats treated with ethanolic extract at doses of 100 and 200 mg/kg showed significant differences at 1 and 3 h from initial levels ($P < 0.001$) (Table 2). The doses of the crude extract produced the maximum glucose lowering (19 and 16%, respectively) in diabetic rats serum and, with the higher dose, a significant time-dependent hypoglycemic effect was shown throughout the period studied.

Based on the result of this study we conclude that the ethanolic extract of *Curculigo orchioides* rhizomes, given orally at a dose of 100 mg/kg b.w., possesses significant hypoglycemic in both normal and glucose loaded rat. Also in diabetic induced rats significantly. It is generally considered that alloxan treatment causes permanent destruction of β-cells (18). The free radical scavenging potential could also help in reducing the known complication of diabetes mellitus (19).

As Venukumar and Latha (2002) reported the antioxidant activity of *Curculigo orchioides*. Thus, the observed antidiabetic effects may be at least partly due to its anti-oxidant activity. Literature review shows that some flavonoids and saponins isolated from medicinal plants significantly reduce the blood glucose

Table 1. Hypoglycaemic activity of Ethanolic extract of *C.orchioides* Gaertn.

Groups	Treatment (po)	Plasma glucose level (mg/dl)		
		0 min	30 min	90 min
I	Control (Distilled water)	75.72±0.49	75.24±0.45	75.12±0.50
II	Extract (100 mg/kg)	73.86±1.22	61.75±1.09 *	58.58±0.83*
III	Glucose (2g/kg)	79.62±1.82	162.5±1.18	134.52±1.67
IV	Glucose + Extract	75.5±0.49	131.56±1.35*	114.9±1.62*
V	Standard (500 µg/kg)+Glucose	74.84±0.92	120.11±2.19*	101.91±1.46*

Values (mg/100ml) are mean ± S.E.M. (n=6), determined at different time (Min) after treatment
A statistically significant difference of the value when compared with the zero time (*p < 0.001).

Table2. Effect of Ethanolic extract of *C.orchioides* on alloxan –induced diabetic rats.

Group	Treatment p.o.	Plasma glucose level (mg/dl)		
		Basal value	1 h	3 h
I	Control(distilled water)	388.54±14.12	379.35±13.34	377.57±12.78
II	EtOh Extract(100 mg/kg)	378.46±13.68	350.78±12.88*	320.36±16.78*
III	EtOh Extract(200 mg/kg)	382.48±16.58	342.92±14.38*	310.64±12.66*
IV	Standard drug Glimeperide (500 µg/kg)	378.58±12.48	338.22±16.92*	308.18±15.46*

Values (mg/100ml) are mean ± S.E.M. (n=6), determined at different time (hour) after treatment
A statistically significant difference of the value when compared with the zero time (*p < 0.001).

levels. (21-26)

Flavonoids Glycosides stimulates the secretion of insulin in β-cells of Pancreas (27). In glucose-loaded animals, it is possible that the extract may act by Potentiation the pancreatic secretion or increasing glucose uptake. On the basis of above evidence it is possible that the presence of glycosides and tannins are responsible for their activity. In conclusion, the ethanolic extracts of rhizome of *Curculigo orchioides* possess a hypoglycemic agent. However, chemical and pharmacological investigations are necessary to identify the latter and to confirm its mechanism of action and its antidiabetic potential.

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