Academic Sciences International Journal of Pharmacy and Pharmaceutical Sciences

ISSN- 0975-1491 Vol 4, Suppl 3, 2012

Research Article

PHARMACOLOGICAL SCREENING OF *SCOPARIA DULCIS* ROOTS FOR HYPOGLYCAEMIC ACTIVITY

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Received: 14 Jan 2012, Revised and Accepted: 02 Mar 2012

ABSTRACT

Present study deals with the pharmacological screening of *Scoparia dulcis* hydro alcoholic extract for its ability to cause reduction in blood glucose levels in experimental rats. The plant material was collected and extracted with hydro alcohol as solvent and the pharmacological screening was carried out at two dose levels that are at 200 and 400 mg/kg using glibenclamide as reference standard in both in alloxan induced and normoglycaemic rats. The test extract has been shown a significant reduction in blood glucose levels at its terminating hour of study that is 8th hour. It was found that test extract has been shown 68.67 and 69.81 percentage reduction in alloxan induced diabetic rats and 19.25 and 24.62 percentage reduction in its blood glucose levels in normoglycaemic rats. The comparable effect of the extract with glibenclamide may suggest similar mode of action, since alloxan permanently destroys the pancreatic β-cells and the extract lowered blood sugar level in alloxanized rats, indicating that the extract possesses extra pancreatic effects. The present research justifies the use of the plant in the folklore diabetic treatments.

Keywords: *Scoparia dulcis,* Glibenclamide, Alloxan monohydrate, Diabetes.

INTRODUCTION

Scoparia dulcis Linn, belonging to family Scrophulariaceae is an erect much-branched herb. It is known as Mithapati in Oriya and Mithipatti in Hindi. The plant is a native of America and widely spread in India. Traditionally, the plant is reported to be used in the treatment of several ailments. The hot aqueous extract of entire plant is reported to be used orally as an aphrodisiac, dysentery, purgative, diabetes, diarrhea, antipyretic, analgesic, gastric disorders and expectorant. The infusion is used externally to treat insect bites, skin wounds and rectally to treat hemorrhoids and roots were employed in diabetes[1-6]. Several phytoconstituents have been reported from the plant earlier. The plant was reported to contain amellin, acacetin, apigenin, cynaroside, linarin, luteolin, vitexin and isovitexin, scutellarein, 7-O-methyl scutellarein and hymenoxin[7]. Presence of diterpenes dulcinol, scoparinol, scopadulcic acid A, scopadulcic acid B, scopadulciol, scopadulin, scoparic acid A, B and C[8]and triterpenes α-amyrin, dulcioic acid, glutinol, ifflaionic acid and betulinic acid have also been reported[9]. Dulcidiol and scopanolal have been reported from the aerial parts[9]. Several pharmacological and biological activities have been reported. The entire plant reported for antimicrobial, hypoglycemic, antispasmodic, analgesic, anticonvulsant, diuretic, cough, anti-inflammatory, antioxidant, anti diabetic, antivenin, antipyretic activities[10, 11]. Even though the plant was reported to have many medicinal uses, there was no scientific literature is available on roots for their hypoglycemic potential. Hence, the present work was taken up to give scientific evidence to the folklore claim on hypoglycaemic activity of roots.

MATERIALS AND METHODS

Plant material

Fresh roots (2 kg) were collected from young matured plants from Eturunagaram, Warangal and authenticated by Botanical survey of India, Howrah. A voucher specimen (Ucpsc/PCOG/KSR/12/2008) has been deposited in the Department of Pharmaceutical Sciences, Andhra University, Visakhapatnam. The collected plant material was shade dried and pulverized.

Preparation of extract

The powdered plant material was extracted with ethanol-water (1:1) by reflux for 8 h. The liquid extract was filtered and concentrated under *vacuum* to yield a dry extract (yield: 11.32 % w/w with respect to dry material) designated as HASD which was used for further study.

Experimental animals

Adult Wistar rats (150-200g) and Swiss albino mice (for toxicity studies) of either sex were used in the studies. The animals were kept in standard polypropylene cages at room temperature of 30 + 2 ^{0}C and 60-65 % relative humidity[12]. All the experimental procedures were approved by Institutional animal ethical committee of Vaagdevi College of Pharmacy, Hanamkonda, Andhra Pradesh, India vide approval No. 1047/AC/09/CPCSEA.

Gross behavioural and toxicity studies

The hydro alcoholic extract of roots of *S. dulcis* was screened for the gross behavioural studies in mice[13, 14]. Adult albino mice of either sex, weighing between 20-25 g were divided into eight groups of six animals each. The control group received 2 ml /kg distilled water orally and other test groups received HASD at dose levels of 200, 400, 800, 1000, 2000, 3000 and 4000 mg / kg in distilled water through oral route. After administration of the dose the animals were observed continuously for first four hours for behavioural changes and for mortality if any at the end of 72 h.

Anti diabetic evaluation

The anti diabetic screening of the hydro alcoholic extract of *Scoparia dulcis* roots was studied in both alloxan induced diabetic and normoglycaemic rats in the following methods.

Using hyperglycaemic rats

The acclimatized animals were kept fasting for 24 h with water *ad libitum* and injected intraperitoneally at a dose of 120 mg/kg of alloxan monohydrate in normal saline. After one hour, the animals were provided feed *ad libitum*. The blood glucose level was checked before alloxanisation and 24 h after alloxanisation by withdrawing blood from the tip of the tail of each rat under mild ether anaesthesia. The blood glucose level was measured with haemoglucostrips supplied by M/s Pulsatum Health Care Pvt. Ltd., Bangalore with the help of a Pulsatum blood glucose monitor.

Animals were considered diabetic when the blood glucose level was raised beyond twice the value of normal. This condition was observed at the end of 48 h after alloxanisation. The animals were

segregated into four groups of six rats in each. Group-I served as control and received vehicle (2 ml/kg) through oral route. Group-II received glibenclamide (2.5 mg/kg). Group-III and IV received the extract at doses of 200 and 400 mg/kg in a similar manner. Blood samples were collected from each rat by cutting the tip of the tail under mild ether anaesthesia. Blood glucose level was estimated at 0 h, 1 h, 2 h,4 h and 8 h respectively[15-17]. The results were expressed as mean ± S.E.M. in Table-1. Significance of difference between control and treated groups was determined using Student's *t*-test.

Using normoglycaemic rats

The animals were fasted for 18 h, but were allowed free access to water before and throughout the duration of experiment. At the end of the fasting period, taken as zero time (0 h), blood was withdrawn from the tip of the tail of each rat under mild ether anaesthesia and the blood glucose was estimated as above. The normal rats were then divided into three groups of six animals each. Group-I served as control and received vehicle (2 ml/kg) through oral route. Group-II received glibenclamide (2.5 mg/kg). Group-III and IV received the extract at doses of 200 and 400 mg/kg in a similar manner. Blood glucose levels were measured after 1, 2, 4 and 8 h of administration of single dose of test samples[18-20]. The results were expressed as mean ± S.E.M. in Table-2. Significance of difference between control and treated groups was determined using Student's *t*-test.

RESULTS

Toxicity studies

The gross behavioural and toxicity studies of the extract revealed no mortality with the tested doses at the end of 72 h. The animals were then kept under supervision up to 14 days after drug administration and no mortality was observed. Hence, HASD was selected for pharmacological screening at dose levels of 200 and 400 mg/kg.

Anti diabetic action

Diabetic rats

In alloxan induced diabetic rats, standard glibenclamide group showed a high significant reduction in blood glucose levels at all the time intervals examined and at 4 to 8th hour, the fall in blood glucose was found to be very good anti diabetic action, where the percentage inhibition in blood glucose level was found to be 72.13 percent at 8th hour (table 1) of study and the blood glucose level was found to be 83.1 and 79 mg/dL at time interval of 4 and 8 hours respectively. In test group, HASD at its two dose levels that is 200 and 400 mg/kg was shown a moderately same action on reduction on blood glucose levels was seen which was supported by its nearly same percentage inhibition at 8th hour, which was found to be 68.57 and 69.81 (table 1) in its two dose levels that is 200 and 400 mg/kg.

Table 1: Result of HASD on blood glucose concentration in alloxan induced hyperglycaemic rats

Group	Treatment	Dose	Blood glucose conc. (mg/dL)				
			0h	1h	2h	4h	8h
I	0.5% w/v Sodium CMC (Vehicle)	2 ml/kg	265.5+11.92	271.8+11.6	275.8+11.47	282.1+11.26	290.8+9.86
II	Glibenclamide	2.5 mg/kg	289.6+22.11	198.1+16.7* (31.59%)	134.8+9.94** (53.54%)	83.1+2.89** (71.29%)	79+2.2** (72.73%)
III	HASD	200 mg/kg	294.8+8.89	261+8.27 (11.47%)	160.8+6.95* (45.45%)	108.6+5.16** (63.14%)	92.6+2.79** (68.57%)
IV	HASD	400 mg/kg	275.5+19.98	208.1+14.42 (24.44%)	118.1+7.61** (57.11%)	96.6+2.13** (64.91%)	83.1+3.44** (69.81%)

Results expressed as Mean + SEM from six observations. Figures in parentheses represent percentage reduction in blood glucose concentration.*P < 0.01, ** P < 0.001

Normal rats

In normoglycaemic rats, standard glibenclamide group showed a moderate reduction of blood glucose levels at time intervals of 4 and 8th hour of study, in which the percentage inhibition was found to be 36.99 percent at terminating hour of the study (table 2) and reduction in blood glucose level was found to be highly significant at 8th hour, which is 62.1 mg/dL. In the test groups HASD at its low dose group that is 200 mg/kg has shown normal level of hypoglycaemic action where as the high dose level it was found to be much interesting action of HASD on blood glucose levels which was found to be 67.3 mg/dL at 8th hour (table 2).

Table 2: Result of HASD on blood glucose concentration in normoglycaemic rats

Group	Treatment	Dose	Blood glucose conc. (mg/dL)				
			0h	1h	2h	4h	8h
I	0.5% w/v Sodium CMC (Vehicle)	2 ml/kg	96.1+3.14	98+3.66	97.1+2.65	96.1+2.06	97.2+2.11
II	Glibenclamide	2.5 mg/kg	98.6+3.6	82.6+2.76* (16.21%)	73.3+2.56** (25.68%)	64.5+1.29** (49.75%)	62.1+0.68** (36.99%)
III	HASD	200 mg/kg	90+4.08	83.5+5.17 (7.22%)	77.5+4.43 (13.88%)	73.6+4.34 (18.14%)	72.6+4.25 (19.25%)
IV	HASD	400 mg/kg	89.3+4.65	84.1+4.21 (5.77%)	80+3.61 (10.44%)	73+2.73 (18.28%)	67.3+4.85* (24.62%)

Results expressed as Mean + SEM from six observations. Figures in parentheses represent percentage reduction in blood glucose concentration.*p< 0.01, **p< 0.001

DISCUSSION

The extract was evaluated for in-vivo anti diabetic activity using alloxan monohydrate at a dose of 120 mg/kg body weight. In the present study it was found that there is a marked elevation in the blood glucose level after administration of alloxan monohydrate. The present studies revealed that the hydro alcoholic extract of *Scoparia dulcis* roots caused significant reduction in the blood glucose levels in the rats. The extract was found to produce marked reduction in blood glucose concentration between 2-4 hours of administration in both alloxan induced hyperglycaemic and normoglycaemic rats at tested dose levels as depicted in table-1 and 2 respectively. When compared with the reference control glibenclamide, the extract caused noticeable reduction in the blood glucose level in both classes of animals except that the onset of action of glibenclamide was noticed from the first one hour.

CONCLUSION

The comparable effect of the extract with glibenclamide may suggest similar mode of action, since alloxan permanently destroys the pancreatic β-cells and the extract lowered blood sugar level in

alloxanized rats, indicating that the extract possesses extra pancreatic effects. The present research justifies the use of the plant in the folklore diabetic treatments. Further study is needed to isolate and characterize the phytoconstituents form *Scoparia dulcis* which are responsible for this potent hypoglycaemic action.

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