

INVIVO SCREENING OF THE LEAF EXTRACTS OF PITHECELLOBIUM

DULCE (Roxb.) Benth. AGAINST DIABETIC SWISS ALBINOMICE

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ABSTRACT

In India, the prevalence of diabetes mellitus is on increase and needs to be addressed appropriately. In this study area, herbal remedies are considered convenient for management of type 2 diabetes with postprandial hyperglycemia due to their traditional acceptability and availability, low costs and lesser side effects. The present study involves screening of **LEAF EXTRACTS OF PITHECELLOBIUM DULCE (Roxb.) Benth** for anti-diabetic activity on alloxan induced diabetic Swiss albino mice using glibenclamide as standard. Ethanol and aqueous extracts (200mg/kg b.w.) of leaves of **PITHECELLOBIUM DULCE (Roxb.) Benth** were screened. Among extracts , alcoholic extracts leaves of **PITHECELLOBIUM DULCE (Roxb.) Benth** shown more significant ($p<0.01$) reduction in blood glucose level in alloxan induced diabetic Swiss albino mice when compared to aqueous .

Keywords: Anti diabetic , blood glucose , glibenclamide , Swiss albino mice

INTRODUCTION

A study of ancient literature indicates that diabetes (madhumeha) was fairly well known and well conceived as an entity in India [3]. The knowledge of the system of diabetes mellitus, as the history reveals, existed with the Indians since prehistoric age. 'Madhumeha' is a disease in which a patient passes sweet urine and exhibits sweetness all over the body, i.e. in sweat, mucus, breathe, blood, etc[4]. The practical usage of juices of various plants achieved the lowering of blood glucose by 10-20%. Diabetes mellitus is one of the common metabolic disorders. Almost 1.3% of the population suffers from this disease throughout the world [1] and number of diabetics is increasing by 6% per year [2]. Approximately 300,000 deaths each year are attributed to diabetes. Its prevalence increases with age,

from about 0.2% in persons less than 17 years of age to about 10% in persons aged 65 years and over. Insulin and oral hypoglycemic agents like sulphonylureas and biguanides [5] are still the major players in the management but there is quest for the development of more effective anti-diabetic agents . *Pithecellobium dulce* Benth. (*Dakhani babul*; *Kodukkapuli*) is used for anti-inflammatory, anodyne, abortifacient, astringent and useful in dysentery, dyspepsia, fever, ulcer, larvicide, sore and venereal diseases. It is a popular drug in the Siddha and Ayurveda systems for treating respiratory and digestive disorders . It also has anti-diabetic activity ,which are efficacious and economical, as compared to synthetic drugs, but not evaluated systematically till date. Hence, the present study was aimed towards the screening of the abovementioned plant extracts for anti-diabetic activity by using alloxan induced diabetic model.

MATERIALS AND METHODS

Plant material

The leaf of *Pithecellobium dulce* were collected from local garden and authenticated at Botany department , wcc , nagercoil.

Plant Extractions

Collected leaves were dried and crushed to coarse powder. Powdered material was charged into soxhlet apparatus and continuous hot extraction was carried out using solvent like ethanol successively water [7].

Animal selection

Healthy adult Swiss albino mice of either sex weighing 150-180 g were selected for the study. The study was carried in accordance with the rules and regulations laid by the Institutional Animal Ethics Committee. The animals were housed with free access to food and water. The basal food intake and body weights to the nearest gram were noted. Mice were starved 24 hr prior to the study. Individual extracts of leafs of were evaluated in five groups of six animals each .

Acute toxicity study

The acute oral toxicity study was carried out as per OECD guidelines. At dose of (2000 mg/kg) 50% mortality was observed. Hence 200 mg/kg b.w. of each extract was taken as effective dose for evaluation of antidiabetic activity [7].

Preparation of doses

All the extracts (200mg/kg b.w.) were suspended in 2% v/w aqueous solution of

glibenclamide (10mg/kg b.w.) as standard drug in normal saline was administered orally. The control group received normal saline orally [8].

Administration of Doses

The test substances were administered in a single dose by gavage using a stomach tube. Animals were fasted 24 hr prior to dosing. During fasting, the animals were weighed and substance was administered. Food was withheld for further 3-4 hr, after the dose administration .

Evaluation of Anti-Diabetic Activity [9, 10, 11]

Before starting the experiment, animals were separated according to their body weight. The animals were injected intraperitoneally (i.p.) at a dose of 150 mg/kg b.w. alloxan monohydrate (sigma chemicals .Inc) freshly prepared in normal saline solution. After one hour of alloxan administration, animals were given feed *ad libitum* and 1ml of (100 mg/ml) glucose i.p. to combat ensuring severe hypoglycemia after 72 hr of alloxan injection; the animals were tested for evidence of diabetes by estimating their blood glucose level using glucometer (Accucheck, Care Pvt. Ltd.,Bangalore) [11] .

To the animals, the test extracts (200 mg / kg b.w. orally) and standard drug glibenclamide tablets (10 mg/kg b.w. orally) were administered by dissolving in 2% water and normal saline respectively[5] . For acute study, 0.2 ml of blood sample was withdrawn through the tail vein puncture technique using hypodermic needle at interval of 0, 2nd, 4th and 6th h of administration of single oral dose. The animals were segregated into seven groups of six rats each for each extract. For all drugs groups of normal, diabetic control and standard glibenclamide were kept same for comparison with alcohol and aqueous extracts of drugs. The mean \pm SEM were statistically calculated for each parameter [10].

RESULTS AND DISCUSSION

The results of effect of extracts of *Pithecellobium dulce* Benth. which are expressed as change in blood glucose level are shown in table . More significant ($p < 0.01$) anti-diabetic activity was observed in alcoholic and aqueous extracts in acute model compared with standard glibenclamide. *In vivo* efficiency was performed in healthy normal Swiss albino mice by measuring the hypoglycemic effect produced after oral administration. Wasan *et al* have suggested that a 25% reduction in blood glucose levels is considered a significant hypoglycemic effect [11]. The results of the study were satisfactory and revealed that the alcoholic extracts of *Pithecellobium dulce* Benth., has exhibited significant hypoglycemic activity. The reduction of blood glucose level in alloxan induced Swiss albino mice was found highest in alcoholic extracts of *Pithecellobium dulce* Benth.

CONCLUSION

The present study involves comparative screening of leaves *Pithecellobium dulce* Benth. for anti-diabetic activity on alloxan induced diabetic Swiss albino mice using glibenclamide as standard. The *in vivo* study demonstrated significant hypoglycemic activity was found highest in alcoholic extracts of *Pithecellobium dulce* Benth.

TABLE 1: EFFECT OF THE ETHANOLIC LEAVES EXTRACTS OF *PITHECELLOBIUM DULCE* (Roxb.) ON BLOOD GLUCOSE OF ALLOXAN INDUCED DIABETIC SWISS ALBINO MICE AFTER ACUTE TREATMENT

Groups (n)	Dose	Initial	Blood	glucose level mg/100ml	(Mean±SEM)
			2nd hour	4th hour	6th hour
Normal control	2ml saline	106±3.27	107±4.91	105±2.66	104±3.06
Diabetic control	2ml saline	281±5.03	290 ±4.61	290 ±2.30	292±3.29
Alcohol extract	200mg/kg b.w.	288±4.09	243±3.24	189±4.10**	186±4.0**
Aqueous extract	200mg/kg b.w.	300±6.18	267±9.08	225.33±7.05**	212 ±5.06**
Glibenclamide	10 mg/kg b.w.	296±4.20	207±1.28	166 ±2.14**	159±1.47**

TABLE 2: EFFECT OF THE AQUOEUS LEAVES EXTRACTS OF *PITHECELLOBIUM DULCE* (Roxb.) ON BLOOD GLUCOSE OF ALLOXAN INDUCED DIABETIC SWISS ALBINO MICE AFTER ACUTE TREATMENT

Groups (n)	Dose	Initial	Blood	glucose level mg/100ml	(Mean±SEM)
			2nd hour	4th hour	6th hour
Normal control	2ml saline	104 ±3.07	107± 2.1	105 ± 2.06	103 ± 3.16
Diabetic control	2ml saline	281±5.03	290 ±4.61	290 ±2.30	292±3.29
Alcohol extract	200mg/kg b.w.	288±4.09	243±3.24	189±4.10**	186±4.0**
Aqueous extract	200mg/kg b.w.	300±6.18	267±9.08	225.33±7.05**	212 ±5.06**

Glibenclamide	10 mg/kg b.w.	296±4.20	207±1.28	166 ±2.14**	159±1.47**

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