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Research Article

ANTIDIABETIC ACTIVITY OF BAUHINIA VARIEGATA EXTRACTS IN ALLOXAN-INDUCED DIABETIC RATS

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ABSTRACT

Bauhinia variegata commonly known as kachnar tree and is cultivated throughout India. Literature survey revealed that the leaf of *Bauhinia variegata* is traditionally used as an astringent in diabetes. Flowers are laxative. The present study was under taken to evaluate the antidiabetic activity of ethanol extract of leaf of *Bauhinia variegata* extract was evaluated using albino wister rat i.e. alloxan induced diabetes in albino wister rat by glucometer method, with 50 mg/kg, 100 mg/kg and 200 mg/kg and higher doses showed significant value represent at table no and figure 1,2,3,4 respectively with different successive extract and show the significant p value.

Keyword: *Bauhinia Variegata*, Ethanolic Extract, Antidiabetic, Alloxan, Albino Wister Rat



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INTRODUCTION

Diabetes mellitus is a group of endocrine syndromes characterized by hyperglycemia; altered metabolism of lipids, carbohydrates, and proteins, and an increased risk of complications from vascular disease¹. Insulin dependent diabetes most commonly afflicts juveniles, but it can also occur in adults. The disease is characterized by an absolute deficiency of insulin caused by massive β -cell lesions or necrosis. Loss of β -cell function may be due to invasion by viruses, the action of chemical toxin or usually through the actions of autoimmune antibodies directed against the β -cells². The metabolic alterations observed are milder than those described for IDDM (for example, NIDDM patients typically are not ketosis) but the long term clinical consequences can be just as devastating (for example, vascular complications and subsequent infection can

lead to amputation of the lower limbs)³. Many remedial plants have been explored and their therapeutic efficacy has been widely exploited in complex disorders such as diabetes⁴. The mountain ebony, *Bauhinia variegata* belongs to the family Fabaceae. It is commonly known as Kachnar in Hindi. Transverse section of the leaf petiole shows single layered epidermis covered with thin cuticle. Covering trichomes showed uni to multicellular broad at the base and pointed at the apex & thin walled multicellular ballon shaped glandular trichomes⁵. The leaf is astringent, tonic and antidiuretic. It is useful in scrofula and skin diseases. It is also used for ulcers and leprosy. The juice of bark is used in the treatment of amoebic dysentery, diarrhoea & other stomach disorders⁶.

The crude extract from *Bauhinia variegata* have been shown to posses biological activities viz., Antihyperlipidemic activity⁷, Antistress/Adaptogenic Activity⁸,

antibacterial, antioxidant⁹, immunomodulatory activities¹⁰, Hepatoprotective activity¹¹, hypoglycemic activity¹². The plant *Bauhinia variegata* has been reported to contain Quercetin, lupeol, β -sitosterol, tannins, kaempferol-3-glucoside, amides, carbohydrates, reducing sugar. Thus, The major objective of this study was to active constituent from the plant and to assess the anti-diabetic potential of *Bauhinia variegata in vivo*¹³.

MATERIAL AND METHOD

Chemicals

Alloxan of CDH, New Delhi was used for the induced of diabetes and was obtained from Department of Pharmacy and the standard drug i.e. glibenclamide was received by Sun Pharmaceutical Industries, Jammu & Kashmir.

Plant material

Fresh and young green leaves of *Bauhinia Variegata* were collected from the vill Gandoli Konch Distt Jalaun (U.P.) and got identified by Dr. R.V. Singh Scientist & Head, Herbarium and Museum Central Council Research Ayurveda & Shidha Gwalior.(M.P.) with (Reference No. CCRAS/10/036).

Preparation of plant extract

Extraction of plant

The freshly collected Leaf *Bauhinia Variegata* were shaded dried until cracking sound was observed during breakage, and then these are made in to coarsely powdered by using dry grinder. The powdered Leaf of the plant (600 gm.) was packed in soxhlet apparatus and continuously extracted with ethanol till complete extraction, after completion of extraction the solvent was removed by distillation and then concentrated extract obtained was dried under reduced pressure using rotatory evaporator at temperature not exceeding 40⁰C and then give moderate heating on water bath. A green extract approximate 450 gm¹⁴.

Anti-diabetic studies

Experimental Animals

The adult male albino rats of weight 180-240 gm were selected for the study. All animals were procured from disease free animal house, Institute of Pharmacy, Bundelkhand University, Jhansi with Institutional ethical number (IAEC No.BU/Pharm/16/01A).

The animals were fasted overnight with water *ad libitum*. The starting dose of 300 mg/kg of methanolic extract was administered orally to three animals in each group. If mortality was observed in two or three animals, then the dose administered was assigned as a toxic dose. If mortality was observed in one animal, then the same dose was repeated again in three animals to confirm the toxic dose. If mortality was not observed, the procedure was repeated for further higher doses such as 200 mg/kg body weight. Animals were observed were observed individually after dosing at least once during the first 30 minutes, periodically during the first 24 hours, with special attention given during the first 4 hours, and daily thereafter, for a total of 14 days, expect

where they need to be removed from the study and humanely killed for animal welfare reasons or were found dead¹⁵.

Preparation of Dose

The Dose of 200 mg/kg and 400 mg/kg of methanol extract was selected for the test. All the doses was given orally after making emulsion in vehicle i.e. 1% acacia gum and the standard drug i.e. glibenclamide was given orally (10 mg/70kg) in the vehicle¹⁶.

The Antihyperglycemic Effect of Ethanolic Extract of Leaves of *Bauhinia Variegata* on Alloxan Induced Diabetic Rats

A) Induction of experimental diabetes:

Diabetes mellitus was induced by administering intraperitoneal injection of alloxan monohydrate 120 mg/kg. to the overnight fasted rats. Fasting blood glucose was determined after depriving food for 16 hrs with free access of drinking water. Hyperglycemia was induced by a single i.p injection of 120 mg/kg of alloxan monohydrate in sterile saline after 2 days of alloxan injection, the hyperglycemia rats (glucose level >250 mg/dl) were separated and divided into different groups comprising of 4 rat each for the anti-diabetic study. The treatment (P.O) was started from the same except normal control and diabetic control group for a period of 10 days. During this period, animal in all group had free access to standard diet and water. Body weight and blood glucose level were estimated on 4th, 7th and 10th day of the treatment¹⁷.

B) Sample collection:

Blood sample were collected from tail nipping and glucose level was determined by an automatic electronic glucometer (Accucheck comfort).

C) Procedure:

After checking the fasting blood glucose in overnight fasted diabetic rats; they were divided into five groups of five rats each and one group of non-diabetic rats.

All the doses were given in the following manner:

- Group A served as normal control and did not receive any treatment.
- Group B served as diabetic control and received alloxan monohydrate and vehicle (0.2 ml of 2% aqueous gum acacia)
- Group C glibenclamide (10 mg / kg p.o) and served as standard
- Group D Ethanolic extract (100 mg / kg p.o)
- Group E Ethanolic extract (250 mg / kg p.o)
- Group F Ethanolic extract (500 mg / kg p.o)

The treatment was continued for 3 hour. During this period, food and water was supplied *ad libitum*. All the doses were administered orally by the oral feeding needle. The effect of extract on blood glucose levels was estimated on overnight fasted rats on hour 0, 1, 2, and 3 by the method described before. The basal values are

those of the day on which extract was started to give. The general behaviors of the animals were recorded

daily. The blood glucose level in (Mean \pm S.E.M.) is shown in the Table 1 and Table 2.

Table 1: The Antihyperglycemic Effect of Ethanolic Extract of Leaves of *Bauhinia Variegata* on Alloxan Induced Diabetic Rats:

Group	Treatment	Fasting blood glucose level (mg/dl)			
		Basal value	4 th day	7 day	10 day
A	Normal control	90.46 \pm 3.80	92.32 \pm 2.92	92.32 \pm 1.73	88.29 \pm 3.44
B	Diabetic control (vehicle)	293.8 \pm 5.27	286.91 \pm 5.05	291.8 \pm 5.41	289.41 \pm 9.75
C	Glibenclamide(10 mg)	285.86 \pm 6.92	205.25 \pm 7.06	183.18 \pm 6.35	178.13 \pm 6.20
D	Ethanolic extract (100 mg/kg)	291.76 \pm 4.79	277.76 \pm 5.65	266.23 \pm 8.19	255.42 \pm 7.71
E	Ethanolic extract (250 mg/kg)	284.48 \pm 5.32	258.23 \pm 6.66	255.85 \pm 9.97	252.06 \pm 9.19
F	Ethanolic extract (500 mg/kg)	287.48 \pm 5.32	212.61 \pm 5.07	198.36 \pm 3.52	189.83 \pm 3.31

Value are mean + SEM, n=6, *P < 0.05, **P < 0.01 and ***P < 0.01 vs diabetic control

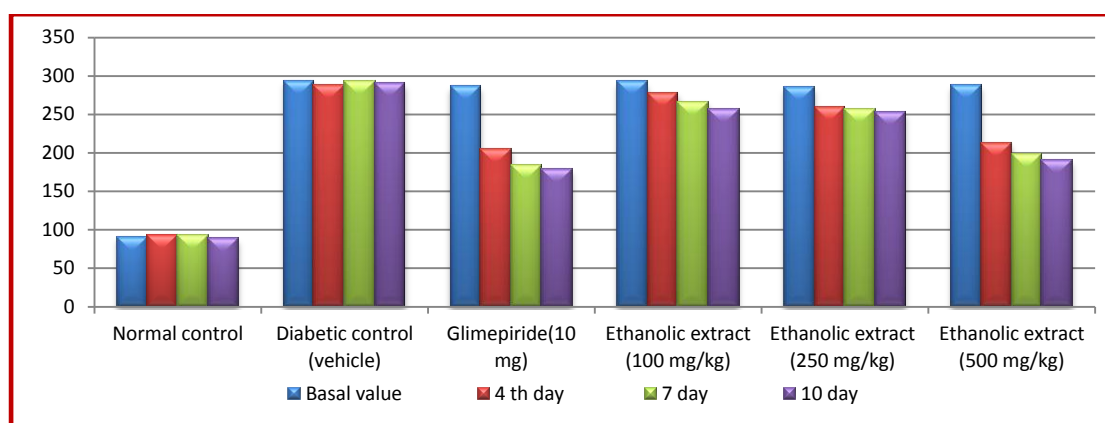


Figure 1: Graphical Comparison of Blood Glucose Level by Different Concentration of Ethanolic Extract of Leaves of *Bauhinia Variegata* And Standard Drug on Alloxan Induced Diabetic Rats

Effect of Ethanolic Extract of Leaves of *Bauhinia Variegata* on oral Glucose Tolerance Test:

The hypoglycemic effect of ethanolic extract of *Bauhinia variegata* leaves was study on glucose loaded rats.

Protocol:

In this glucose tolerance test fasted five groups of five animals each group 1 served as control and received vehicle. Group 2 received standard drug glibenclamide at on i.p dose of 120 mg / kg and group 3, 4 and 5

received ethanolic extract orally at a dose of 100 mg/kg, 250 mg/kg and 500 mg/kg. The rats of the entire group were given glucose (2g/kg), 30 min after the extract and drug administration. Blood samples were collected by tail nipping just prior to glucose administration 0 hour, 30minutes, 60minutes and 120minutes after glucose loading and blood glucose level were measured by glucometer. Basal values are those after which glucose was administrated.

Table 2: The Antihyperglycemic Effect of Ethanolic Extract of Leaves of *Bauhinia Variegata* on Glucose Loaded Rats:

Group	Dose mg/kg	Blood glucose level (mg/dl)			
		0 min	30 min	60 min	120 min
1	Control (2mg/kg)	78.2 \pm 3.20	168 \pm 3.5	143.8 \pm 2.37	130 \pm 1.84
2	Glibenclamide (120mg/kg)	76.5 \pm 2.45	145.2 \pm 3.35**	111.5 \pm 3.11**	82.4 \pm 2.40**
3	<i>Bauhinia variegata</i> Ethanolic extract (100 mg/)	72.2 \pm 2.37	152.2 \pm 1.77**	136.2 \pm 1.76 (NS)	124.2 \pm 2.26 (NS)
4	<i>Bauhinia variegata</i> Ethanolic extract (250 mg/)	73.3 \pm 3.23	157.3 \pm 2.56	130.2 \pm 3.26	114.8 \pm 3.05
5	<i>Bauhinia variegata</i> Ethanolic extract (500 mg/)	79.3 \pm 3.30	150.8 \pm 2.64**	122.4 \pm 2.40**	90.5 \pm 1.70**

NS= not significant, **P < 0.01, show significant when compare with control

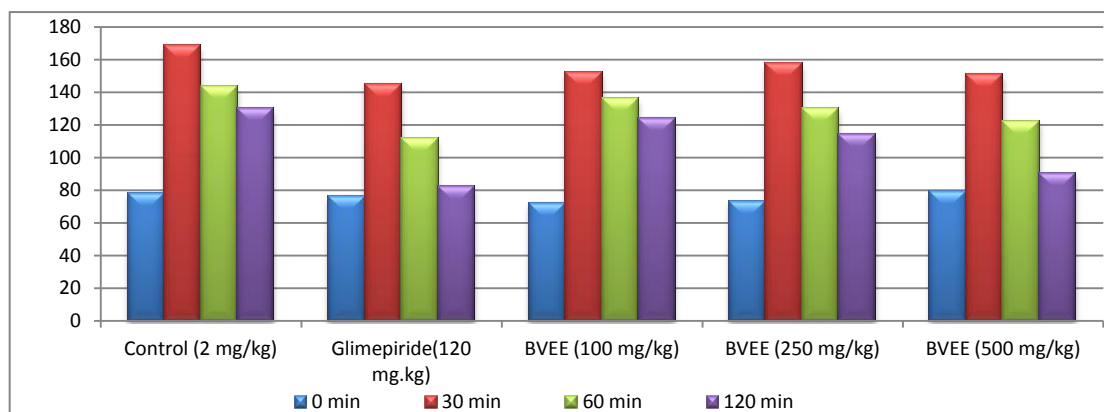


Figure 2: Graphical Comparison of Blood Glucose Level by Different Concentration of Ethanol Extract of Leaves of *Bauhinia Variegata* And Standard Drug on Glucose Loaded Rats:

Statistical Analysis

The data were statistically evaluated using one way Anova. expressed as Mean \pm S.E.M. followed by Tukey test using the Graph pad instant Demo (Data set 1.IS) version P. values of 0.05 or less were considered to be significant.

RESULT AND DISCUSSION

Ethanol extract of bauhinia variegata leaves was subjected to antidiabetic activity in rats where alloxan monohydrate (120 mg/kg, b.w, i.p) used as the diabetogenic agent. A marked rise in fasting blood glucose level observed in diabetic control compare to normal control rates. Ethanol extract of bauhinia variegata (at 250 and 500 mg/kg) exhibited a dose dependent significant anti-hyperglycemic activity on 4th, 7th, and 10th day part treatment. The extract dose of 100 mg/kg also caused reduction in blood glucose level but the result was found statistically insignificant. The

antihyperglycemic effect of ethanol extract at was found less effective than the reference standard. Glibenclamide produced a significant reduction in blood glucose compare to diabetic control. When the activity of extract was done by glucose tolerance test in glucose loaded rats ethanol extract should significant effect on the blood glucose level but extract of 100mg/kg did not show the significant result. ethanol extract 250mg/kg and 500mg/kg showed the significant decrease in blood glucose level.

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