

Research Article

Antidiabetic Activity of Methanolic Extract of Hibiscus Mutabilis Leaves Against Alloxan Induced Diabetes in Rats

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ABSTRACT

Diabetes is chronic diseases spread worldwide in all type of age group. This research work show anti-diabetic activity of *Hibiscus mutabilis* in the form of methanolic extract. The methanolic extract of leaf of *Hibiscus mutabilis* (Malvaceae) were tested for anti-diabetic activity in alloxan induced diabetes wistar rats and compared with standard. Methanolic extract has shown significant protection and maximum reduction in glucose. The maximum reduction in glucose was observed after 6 hours at a dose level of 200 mg/kg of body weight. Blood sugar level was determined using digital glucometer. The results of this comprehensive study reveal that *Hibiscus mutabilis* leaf shown statistically significant anti-diabetic activity in comparison to the standard glibenclamide.

Keywords: *Hibiscus mutabilis*, anti-diabetic, alloxan, glibenclamide.

INTRODUCTION

Diabetes is one of the most prevalent chronic diseases in the world¹. By the year 2025, India shall have the maximum number of diabetes in the world making it, the "Diabetes capital of the world"². Diabetes mellitus (DM) is a metabolic disorder characterized by hyperglycemia resulting from defects in insulin secretion, insulin action or both³. More than 400 species of plants have been reported to display hypoglycemic effects, but only a few of them have been investigated. There are only a few reports on the effects of this plant in the literature and some of them presenting contradictory or unsuccessful results. Presently there are different groups of oral hypoglycemic agents for clinical use, having characteristic profiles of side effects⁴. Management of diabetes without any side effects is still a challenge to the medical system. This leads to increasing demand for natural products with antidiabetic activity having fewer side effects. Type-1 diabetes is an autoimmune disease. The most common form of diabetes is type-2 diabetes. About 90 to 95 % of people with diabetes have type-2. Non insulin dependent diabetes mellitus usually occurs after the age of forty⁵. This form of diabetes is associated with older age, obesity, family history of diabetes, previous history of gestational diabetes, physical inactivity and ethnicity. About 80% of people with type 2 diabetes are overweight⁶. The use of ethnobotanicals has a long folkloric history for the treatment of blood glucose

abnormalities⁷. Therefore, the search for more effective and safer hypoglycemic agents has continued to be an important area of active research.

MATERIALS AND METHODS

Plant Material

Hibiscus mutabilis leaves were collected from south 24 parganas district, West Bengal, India and identified from authentic sources. The collected leaves were washed thoroughly with tap water and dried in shade, crushed to coarse powder and used for further studies.

Preparation of extract

100gm. of powdered leaves were extracted with 80% methanol using continuous hot extraction method. The extracts were evaporated to dryness. The dried extract of *Hibiscus mutabilis* thus obtained was used for the assessment of antidiabetic activity.

Animals

Albino wistar male rats weighing 150-200g was used for the present study. The animal house was well ventilated and animals had 15-20 ± 2°C. The animals were housed in large spacious hygienic cages during the course of the experimental period. The animals were fed with rat pellets feed supplied by M/s Hindustan Lever Limited, Bangalore, India and filtered water ad libitum. Animals described as fasted were deprived of food for ≥16 hr but allowed free access to water. The place where the experiments were conducted was kept very

hygienic by cleaning with antiseptic solution, as the diabetic animals are susceptible to infections⁸. All the studies conducted were approved by the Institutional Animal Ethical Committee (IAEC) of Malhotra College of pharmacy, Bhopal.

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The extract of *H. mutabilis* was found to be safe for further biological studies as no toxic effect and lethality was observed up to 3000 mg/kg per oral in rat. Only the consumption of food was increased by 20% in the dose of 2000 and 3000 mg/kg during 4h but remaining normal afterwards.

Experimental design

Albino Wistar rats (180-230g) of either sex were randomly divided in to 6 groups (6 rats/group) and were fasted overnight (18hrs). Animals in-group I were treated with acacia (5%) as control, remaining groups animals were treated with freshly prepared aqueous solution of alloxan monohydrate in a dose of 150mg/kg body weight through intraperitoneal route^{9,10}. Then 5% dextrose was administered to combat the immediate hypoglycemia. Group II kept as a diabetic control. After 18 hrs, Group III animals treated with standard glibenclamide (600µg/kg b.w) through oral route, even Group IV, V, VI and animals were treated with 50, 100 and 200 mg/kg of leaf of *H. mutabilis* of methanolic extract respectively through oral route. Blood samples were taken from the tail vein at 0, 1, 2, 3, 4, 5 and 6 hrs. The blood glucose concentration was measured by using glucometer and noted¹¹.

Statistical analysis

The data was statistically analyzed by one-way ANOVA followed by Dunnett multiple comparison test with equal sample size. The difference was observed as significant when $p < 0.01$. All the values were expressed as mean \pm standard deviation (S.D.). Glucose level was observed at dose of 200 mg/kg of aqueous methanol extracts of leaf of *H. mutabilis*.

RESULTS AND DISCUSSION

Based on the acute toxicity studies, the dose of leaves extracts was selected for further studies. Table 1 represents the evaluation of antidiabetic activity of *H. mutabilis* leaves methanolic extracts (50, 100 and 200mg/kg) and glibenclamide fed alloxan induced diabetic wistar rats shown the changes in the levels of blood glucose. Methanolic *H. mutabilis* leaf extract has shown maximum reduction in blood glucose level which calculated by comparing the blood glucose level at 6th hr with the blood glucose level at 0 hr of its respective groups based on the dose. The finally percentage reduction of blood glucose represented 200mg/kg concentrated methanolic extract has shown maximum reduction in blood glucose as compared to control than other concentrations of *H. mutabilis* leaf extracts. Group II, III, IV, V and VI showed suppression of blood glucose level at 6 hrs significantly ($p < 0.01$) compared to zero hour to its respective group. In this study, 200mg/kg of methanolic *H. mutabilis* leaf extract significantly ($p < 0.01$) suppressed blood glucose. Alloxan, a beta cytotoxin, destroys β -cell of islets of Langerhans of pancreas resulting in a decrease in endogenous insulin secretion and paves ways for the decreased utilization of glucose by body tissues¹². From the study, it is suggested that the possible mechanism by which the plant extract decrease the blood glucose level may be by potentiation of insulin effect either by increase in pancreatic secretion of insulin form beta cells of islets of langerhans.

CONCLUSION

In the present study, *Hisiscus mutabilis* showed portent antidiabetic effect in alloxan-induced diabetic rats and reduced the mortality rate significantly. The present investigation has also opened avenues for further research, especially with reference to the different dose studies and development of potent formulation for DM from *Hisiscus mutabilis*. Activity guided fractionation, formulation, and its evaluation will be a need for the future study.

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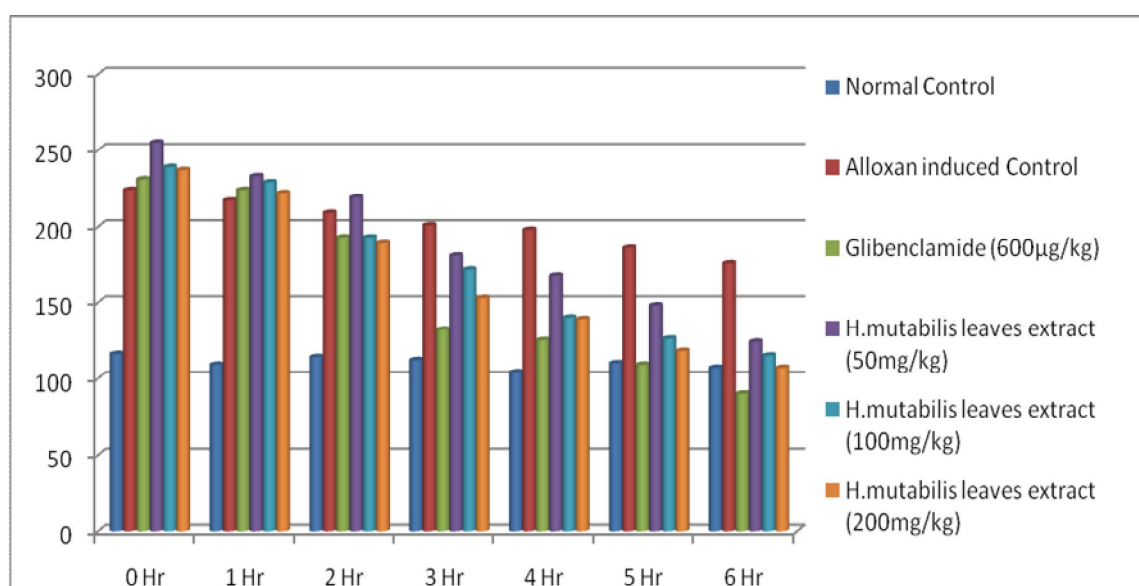
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Table 1: Anti diabetic activity of methanolic *Hisiscus mutabilis* leaves extracts

Interval	Group I	Group II	Group III	Group IV	Group V	Group VI
	Normal Control	Alloxan induced Control	Glibenclamide (600µg/kg)	H.mutabilis leaves extract (50mg/kg)	H.mutabilis leaves extract (100mg/kg)	H.mutabilis leaves extract (200mg/kg)
0 hr	116±4.02	224±5.65	231±8.02*	255±.24	239±6.12	237±0.01*
1 hr	109±0.09	217±3.6	224±0.21*	233±5.56	229±4.62	222±2.4*
2 hr	114±7.05	209±6.11	193±9.56**	219±4.71	193±2.97	189±8.34
3 hr	112±5.73	201±6.66	132±1.07**	181±7.09*	172±4.98	153±5.86
4 hr	104±3.85	198±7.45	125±9.01**	168±5.23	140±2.84	139±4.90*
5 hr	110±6.90	186±6.87	109±3.83**	148±2.71	126±4.06	118±6.03
6 hr	107±5.71	176±7.75	90±2.06**	124±1.08*	115±6.93*	107±4.09*

n=6; values expressed as mean ± S.D

* p<0.01, ** p<0.001

**Graph 1: Anti diabetic activity of methanolic *Hisiscus mutabilis* leaves extracts****REFERENCES**

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