Original Article

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ANTI-HYPERGLYCEMIC ACTIVITY OF *HYGROPHILA SPINOSA* ROOTS IN ALLOXAN-INDUCED DIABETIC RATS

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Abstract

The objective of this study was to verify the claims and to evaluate the anti-diabetic activity of the roots of Hygrophila spinosa, by preparing the various organic extracts and the resultant extracts are to be screened for the anti-diabetic activity to prove the claimed activity. The chloroform, ethyl acetate and alcohol extracts of Hygrophila spinosa, roots were prepared and subjected for phytochemical screening and tested for their anti-diabetic activity on alloxan-induced diabetic rats. Phytochemical screening showed positive test for steroids and/or triterpenoids ($CHCl_3$ extract), steroids and/or triterpenoids and their glycosides (EtoAc extract), steroids and/or triterpenoids and their glycosides (EtOH extract). The ethyl acetate and ethanol have shown significant anti-diabetic activity at a dose of 200 mg/kg, p.o. The study results suggested that Hygrophila spinosa, roots possessed potential anti-diabetic activity.

Key words: *Hygrophila spinosa*, Anti-Hyperglycemic activity, Alloxan.

Introduction

Hygrophila spinosa (Acanthaceae) is commonly found in water-logged areas throughout India[1]. The plant is used as a diuretic and for the treatment of rheumatism, jaundice, inflammation, pain, hepatic obstruction, gout, bacterial infection etc[2-6]. The aerial parts of the plant are reported to contain lupeol, stigmasterol and butelin while the seeds mainly contain fatty acids. Its root contains an alkaloid named hygrosterol[7] while its flower contains apigenin 7-O-glucuronide[8-9]. However, no data were found regarding the pharmacological and phytochemical evaluation of the roots of the plant. The aim of the present study is to investigate the Antihyperglycemic activity of various ($CHCl_3$ EtoAc and EtOH) extracts of the roots of *H. spinosa*.

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Materials and Methods

Plant material

The roots of *Hygrophila spinosa* were collected from Bhimavaram, West Godavari District, Andhra Pradesh, India, and was authenticated by Professor K. Venkiah, Dept. of Botany, Andhra University, Visakhapatnam. A voucher specimen has been deposited at the museum of our college. After collection, the roots were washed thoroughly under running tap water, cut into pieces , shade dried at room temperature (24-26^{0}C) and ground into a coarse powder.

Extraction and isolation

The coarse powder of Hygrophila *spinosa roots* (2.5kg) was extracted successively with chloroform, ethyl acetate and alcohol in a Soxhlet extractor and yield was 0.19, 0.75 and 0.39%, respectively, on dried weight basis.

Phytochemical screening of organic extracts

Freshly prepared organic extracts were tested for the presence of alkaloids, steroid and/or triterpenoids and their glycosides, tannins, flavonoids and their glycosides, carbohydrates and cardiac glycosides using standard procedure[10].

Test animals

Male Wistar rats weighing 170-210 gms were used in the experiment. They were maintained in standard laboratory conditions of temperature (25 ± 2°C), relative humidity (55±10%) and 12 h dark/light cycle were used. They were fed with standard diet (Hindustan Lever Ltd, India) and water *ad libitum*.

Antidiabetic activity

Anti-diabetic activity of various organic extracts was studied in alloxan-induced diabetic rats. Hyper -glycemia was induced by a single *intraperitoneal* injection of 120 $mg.kg^{-1}$ of alloxan monohydrate in sterile saline[11]. After 5 days of alloxan injection, the diabetic rats (glucose level >350 $mg.dl^{-1}$) were separated and divided into five groups of six animals each (Table 01). Group I served as diabetic control and was given distilled water. Groups II-IV were treated orally with chloroform ($CHCl_3$), ethyl acetate (EtoAc) and alcohol (EtOH) extracts at a dose of 200 $mg.kg^{-1}$ suspended in carboxy methylcellulose (0.25%). The standard anti-diabetic drug, Tolbutamide was administered orally at a dose of 50 $mg.kg^{-1}$ to Group-V animals. Blood samples were collected from the tail vein at designated time points after extracts and standard drug administration. Plasma was harvested and blood glucose levels were measured immediately by glucose oxidase method[12].

Table 01: Test Design for Activity

Group	Treatment	Rat number
I	Diabetic rats – Untreated	6
II	Diabetic rats treated with 200 $mg{\cdot}kg^{-1}$ of Chloroform Extract	6
III	Diabetic rats treated with 200 $mg{\cdot}kg^{-1}$ of Ethyl acetate Extract	6
IV	Diabetic rats treated with 200 $mg{\cdot}kg^{-1}$ of Alcohol Extract	6
V	Diabetic rats treated with 50 $mg{\cdot}kg^{-1}$ of Tolbutamide	6

Table 02: Effect of *Hygrophila spinosa* Roots Extracts on Fasting Blood Glucose levels in Diabetic rats

Group	0 h	2 h	4 h	6 h
I	376.01 ± 6.75	371.45 ± 7.02	366.21 ± 7.64	361.01 ± 6.74
II	379.14 ± 7.06	394.52 ± 7.00	367.22 ± 7.54	360.71 ± 7.32
III	396.07 ± 7.85	360.24 ± 8.03**	278.25 ± 7.40***	289.22 ± 7.37***
IV	512.00 ± 7.06	500.34 ± 14.75	423.34 ± 18.60**	354.00 ± 3.70***
V	444.27 ± 8.70	364.41 ± 14.30***	288.23 ± 5.07***	201.54 ± 2.54***

Values (mg 100 ml^{-1}) are mean ± S.E (n = 6), determined at different time (h) after treatment.
*p < 0.05; ** p < 0.01; ***p < 0.001 v_s 0 h; Student's *t*-test

Results and Discussion

Phytochemical screening gave positive test for triterpenoids and steroids ($CHCl_3$ extract), steroids and/or triterpenoids and their glycosides (EtoAc extract), steroids and/or triterpenoids and their glycosides (EtOH extract). The $CHCl_3$ extract of *Hygrophila spinosa*, roots did not show any antihyperglycemic activity. On the contrary, the EtoAc extract after oral administration of 200 $mg{\cdot}kg^{-1}$ exhibits very significant reduction in blood glucose levels. Similarly, the EtOH extract has produced significant reduction in blood glucose levels at 200 $mg{\cdot}kg^{-1}$. As shown in Table 02, the effect of EtoAc and EtOH could be comparable to that of well-known hypoglycemic compound, Tolbutamide (50 $mg{\cdot}kg^{-1}$). Thus the claim made by the traditional Indian system of medicine regarding the use of roots of this plant in the treatment of diabetes stands confirms. As far as the mechanism of action is concerned, we can predict that the antihyperglycemic activity of *Hygrophila spinosa could* be due to an enhancement of peripheral metabolism of glucose, even if an increase of insulin

release cannot be excluded. Present evaluation work is directed to isolate the active principles from EtoAc and EtOH extracts of *Hygrophila spinosa*, *roots* and elucidation of mechanism of action.

References

1. The Ayurvedic Pharmacopoeia of India. Part I, vol. II,st edn., Govt. of India, Ministry of Health &Family Welfare, Department of ISM & H, Delhi,The Controller of Publications, 1999, 88-94.
2. Sharma PC, Yelne MB, Dennis TJ. Database onmedicinal plants used in ayurveda. vol. 4, NewDelhi, Central Council for Research in Ayurveda & Siddha, 2002, 320-331.
3. Chopra RN, Nayar SL, Chopra IC. Glossary of IndianMedicinal Plants. New Delhi, CSIR, 1986, 29.
4. Nadkarni KM. Indian Materia Medica. Bombay, India,Popular Prakashan, 1978, 667-669.
5. Mazumder UK, Gupta M, Maiti S, Mukherjee D.Antitumor activity of *Hygrophila spinosa* onEhrlich ascites carcinoma and sarcoma- 180 induced mice. Indian J Exp Biol., 1997; 35: 473-477.
6. Boily Y, Vanpuyvelde L. Screening of medicinal plants of Rwanda (Central Africa) for antimicrobial activity. J Ethnopharmacol., 1986; 16: 1-13.
7. Quasim C, Dutta NL. Chemical investigation of *Asteracantha longifolia* Nees. J Indian Chem Soc., 1967; 44: 82-83.
8. Usha K, Kasturi GM, Hemlatha P. Hepatoprotective effect of *Hygrophila spinosa* and Cassiaoccidentalis on carbon tetrachloride induced liver damage in experimental rats. Indian J Cli.Bio.chem., 2007; 22: 132-135.
9. Balraj P, Nagaraj S. Apigenin 7-oglucuronide from the flowers of *Asteracantha longifolia* Nees.Indian Drugs., 1982; 19: 150-152.
10. Farnsworth NR. Biological and phytochemical screening of plants. *J Pharm Sci.,* 1966; 55(3): 225-276.
11. Joy KL, Kuttan R. Anti-diabetic activity of *Picrorrhiza kurroa* extract. *J Ethnopharmacol.,* 1999; 67(2): 143-148.
12. Trinder P. Determination of glucose using glucose oxidase with an alternative oxygen receptor. *Ann Clin Biochem.,* 1969; 6(1): 24-27.