

Hypoglycemic activity of aqueous extract of *Urtica parviflora* roxb. in normoglycemic rats

Sangeeta Pilkhwal Sah^{1*}, Mukesh Lal Sah¹, Vijay Juyal¹, Savita Pandey¹

*Corresponding author:

Sangeeta Pilkhwal Sah
¹ Department of
Pharmaceutical Sciences,
Bhimtal Campus, Kumaun
University Nainital,
Uttarakhand, 263136, India
E-mail:
spilkhwal@rediffmail.com

Abstract

In the present study aqueous and ethanolic extract of leaves of *Urtica parviflora* were evaluated for hypoglycemic effect in normal rats using both 18 hr fasted rat model and oral glucose tolerance test. The aqueous extract of leaves showed a good hypoglycemic response in both the models, while ethanolic extract exhibited very weak but insignificant effect, only in 18 hr fasted rat model. The aqueous extract was further tested for effect on intestinal glucose absorption. The amount of glucose absorbed in a segment of jejunum in situ was 13 ± 0.75 mg in presence of aqueous extract vs. vs. 9.05 ± 0.68 mg in control rats during 2 h ($P < 0.05$). Phytochemical screening of aqueous extract revealed the presence of alkaloids, reducing sugars, polysaccharides, tannins, saponins, glycosides and flavonoids. The results indicate that aqueous extract possess significant hypoglycemic activity which may be attributed to, in part by reduction of intestinal glucose absorption by the abovementioned chemical constituents.

Keywords: Hypoglycemic activity, *Urtica parviflora*, Oral glucose tolerance test

Introduction

Urtica is a genus of annual or perennial herbs, commonly known as Nettle distributed in the temperate and subtropical zones, armed with stinging hairs on the leaves and stems which on contact with the skin, cause irritation and symptoms of urticaria and nettle rash. Acetylcholine, histamine and 5-hydroxytryptamine have been implicated in urticaria and itching from the stinging hairs. Four species occur in India of which *Urtica parviflora* Roxb. is found abundantly in Kumaon and Garwhal between 1,500 ft to 7000 ft elevation. Young branches and leaves of *U.*

parviflora are used as delicious pot herbs. Seed oil is edible as well as medicinal in sciatica, rheumatism and several skin ailments; hair wash from leaf extract is believed to avoid baldness. The leaves are used in dysentery, joint pain and liver disorders [1]. The roots are employed for the treatment of fractures of bone and dislocations of joints [2]. Despite several experimental studies on other *Urtica* species [3-8], there is currently only one literature depicting protective effect of *U. parviflora* Roxb. in CCl₄ hepatotoxicity [9]. Studies on hypoglycemic [10, 11] and antihyperglycemic

[12] effects have been carried on other species like *U. pilulifera* and *U. dioica*. With this background present study was designed to evaluate hypoglycemic activity of *U. parviflora* Roxb. in normoglycemic rats.

Materials and methods

Plant identification

The leaves of *Urtica parviflora* Roxb. was collected in the month of June from Nainital and was identified from Botanical Survey of India, Dehradun. A voucher specimen (No. 112286) has been kept in Department of Pharmaceutical Sciences, Kumaun University, Nainital, India.

Preparation of extracts

The collected leaves were dried at room temperature and 20 g was boiled with 200 ml of distilled water for 20 min with an occasional stirring. The extract was evaporated in vacuum to give a crude residue (yield: 10.6%). Similarly 20 g of leaves was macerated with 50% ethanol for 24 hrs. The ethanolic extract obtained was concentrated in rotavapor at 60°C to give crude residue (yield: 7.6%).

Animals

Male Wistar rats weighing 250–350 g of both sexes were kept in a room maintained at a temperature of 23±2 °C and under 12 h light/dark cycle.

Hypoglycemic activity

Both the extracts of *Urtica parviflora* viz. aqueous extract of leaves (UPA) and 50 % ethanolic extract of leaves (UPE) were evaluated for the hypoglycemic activity using following experimental models.

18h fasted rat model

Animals were divided into 5 groups each containing 6 animals. Grouping was done as:

Group I- Control group: 1ml/kg distilled water p.o.

Group II- UPA: 250 mg/kg p.o.

Group III- UPE: 250 mg/kg p.o.

Group IV- Glibenclamide 2 mg/kg p.o.

All the animals were fasted for 18 hrs and then blood glucose level was determined immediately before treatment and then 3 hrs after treatment.

Oral glucose tolerance test (OGTT)

Animals were fasted for 16 h before the OGTT. Glucose (1 g/kg) was administered by gavage 30 min after oral administration of 250 mg/kg of *U. parviflora* extracts. Glibenclamide at dose of 2 mg/kg was used as a reference drug. Blood glucose level was measured each hour after glucose loading in rats under light ether anesthesia. Blood was obtained from tail vein using heparinized capillary tubes and immediately centrifuged for 5 min. Plasma was analyzed for glucose content using a glucose oxygenase method (Sigma diagnostics).

Blood sampling

Approximately, 1 ml blood sample obtained from each animal was placed into an Eppendorf tube, centrifuged at 3000 rpm. The sera were collected and the blood glucose was estimated by glucose oxidase method [13].

Intestinal glucose absorption

The effect of the aqueous extract (UPA) was tested in a perfused jejunum segment (6 cm) in fasted rats for 36 h and anaesthetized with sodium pentobarbital (50 mg/kg, i.m.). The composition of the perfusing solution was as follows: 126 NaCl, 2.7 KCl, 0.4 NaH₂PO₄.6H₂O, 1.0 MgCl₂.6H₂O, 6.9 CaCl₂, 7.1 NaHCO₃ and 5.6 glucose mM, at pH 7.5. The aqueous extract was added to a final concentration of 250 mg/kg. The system was set at constant temperature of 37°C, and the perfusion rate was 0.53 ml/min for 2 h [12]. The controls were perfused with the perfusing solution without the extract.

Phytochemical screening

Preliminary phytochemical screening of aqueous extract of *U. parviflora* leaves, was done for their chemical constituents [14].

Following reagents and chemicals were used. Alkaloids with dragendroff's reagents, flavonoids with metallic magnesium plus HCl, saponins with the ability to produce foam, reducing sugars with Fehling's reagent, glycosides with Liberman's test, tannins with ferric chloride and polysaccharides with iodine solution.

Statistical Analysis

All data were expressed as mean \pm SEM of 6 animals. Results were analysed statistically by One-way Analysis of Variance (ANOVA) followed by Tukeys multiple comparison using sigma stat software. Values of $P < 0.05$ were considered statistically significant.

Results and Discussion

The results of the present study demonstrated that the aqueous extract (UPA) of *U. parviflora* had a significant hypoglycemic effect in normoglycemic rats. The results showed that in fasted rats aqueous extract of leaves (UPA) produced an 18 mg/dl fall in blood glucose level after 3 hr of treatment, however there was a weak but insignificant decrease (12 mg/dl) in blood glucose level in the group treated with 50% ethanolic extract of leaves (UPE) (Table-1). In the control group blood glucose level increased by 4 mg/dl while group receiving glibenclamide showed a 32 mg/dl decrease in blood glucose level.

In oral glucose tolerance test, only aqueous extract was found to be active comparable to that of the glucose treated control group (Fig. 1). A good antihyperglycemic effect of UPA was observed at the first hour after glucose loading in rats under OGTT (Fig. 1). The fall of glycemia was approximately 22 % vs. control. This effect was still present 180 min after the oral administration of glucose. Glibenclamide at dose of 2 mg/kg resulted in a significant decrease in glucose level at all the time interval with the exception of the effect observed 1 h after glucose loading. The aqueous extract UPA which exhibited significant hypoglycemic and

antihyperglycemic was evaluated for effect on intestinal glucose absorption. The amount of glucose absorbed in controls during 2 h was (13 \pm 0.75 mg) vs. (9.05 \pm 0.68 mg) in the presence of aqueous extract ($P < 0.05$).

Table 1. Acute effect of the extracts from *Urtica parviflora* on blood glucose levels in normal rats.

Treatment (mg/kg, p.o.)	Blood Glucose (mg/dl) Mean \pm S.E.M	
	0 h	3 h
Glibenclamide (2)	81 \pm 3.2	49 \pm 2.9 * (39)
UPA (250)	87 \pm 2.8	69 \pm 3.4* (20)
UPE (250)	86 \pm 3.4	74 \pm 3.0 (13)
Control	75 \pm 3.6	79 \pm 2.4

Values are mean \pm S.E.M. for n=6. Data analysis was performed using one way ANOVA followed by Tukeys test. * $P < 0.05$ vs. control.

The extract did not produce any intestinal irritation. The results showed that water extract significantly reduced the absorption of glucose which can be considered as one of the mechanisms by which this extract can regulate the glucose homeostasis in glucose loaded rats. Phytochemical screening of aqueous extract UPA showed the presence of alkaloids, polysaccharides, reducing sugars, saponins. In many studies alkaloids have been found to have hypoglycemic activity by inhibiting glucose transport through the intestinal epithelium [15]. Hypoglycemic plants are found to contain polysaccharides and the various experimental results indicate that the polysaccharides increase the levels of serum insulin, reduce the blood glucose levels and improve tolerance of glucose [16]. Saponins [17] and flavonoids [18] are also known to possess potent hypoglycemic activity. As a conclusion, it could be speculated that the observed hypoglycemic and antihyperglycemic activity of *Urtica parviflora* leaves might be

related to the presence of alkaloids, polysaccharides, saponins and flavonoids.

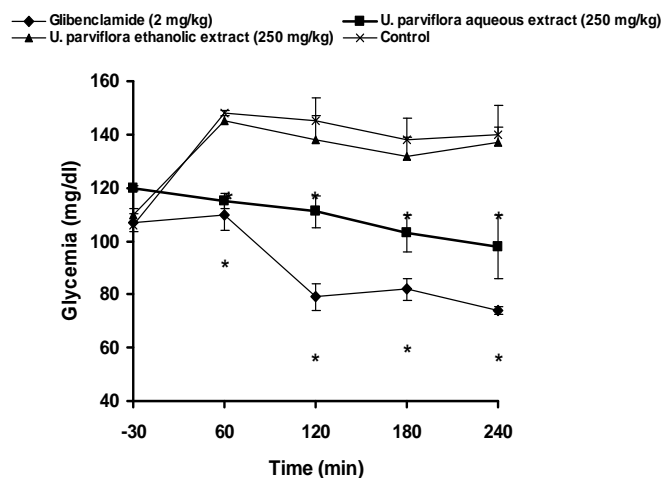


Figure 1. Effect of oral administration of 250 mg/kg of *U. parviflora* extracts on plasma glucose level of oral glucose loaded normal rats. Values are means±S.E.M. * P<0.05 vs. control (n=6).

Conclusion

The result depicted that aqueous extract of leaves of *Urtica parviflora* has good hypoglycemic and antihyperglycemic effect and the primary action being inhibition of intestinal glucose absorption. The study is only preliminary and based on the present findings; antidiabetic potential of the extract can further be studied in diabetic rats. In addition the extract could further be subjected to bioactivity guided drug discovery to isolate lead compounds that are responsible for such activity and other experiments are necessary to determine the other mechanisms responsible for the abovementioned effects.

References

- Gurung G. The Medicinal Plants of Sikkim Himalaya. Ist ed. Subhash Publication, Sikkim; 1999.
- Ramachandran K. Wealth of India (Raw Materials). Publications and Information Directorate, Council of Scientific and Industrial Research, New Delhi; 1992.

- Gulcin I, Kufrevioglu OI, Oktay M, Buyukokuroglu ME. Antioxidant, antimicrobial, antiulcer and analgesic activities of nettle (*Urtica dioica* L.). J Ethnopharmacol 2004; 90: 205-215.
- Yongna Z, Wantana R, Pisit B, Zhongkun L, Rongping Z. Analgesic and antipyretic activities of the aqueous extract of *Urtica macrorrhiza* in experimental animals. Fitoterapia 2005; 76:91-95.
- Kanter M, Coskun O, Budancamanak M. Hepatoprotective effects of *Nigella sativa* L and *Urtica dioica* L on lipid peroxidation, antioxidant enzyme systems and liver enzymes in carbon tetrachloride-treated rats. World J Gastroenterol 2005; 11:6684-6688.
- Obertreis B, Giller K, Teucher T, Behnke B, Schmitz H. Anti-phlogistic effect of *Urtica dioica* folium extract in comparison to caffeoyl malic acid. Arzneimittel Forschung 1996; 46: 52-56.
- Akbay P, Basaran AA, Undeger U, Basaran N. In vitro immunomodulatory activity of flavonoid glycosides from *Urtica dioica* L. Phytother Res 2003; 17:34-37.
- Legssyer A, Ziyat A, Mekhfi H, Bnouham M, Tahri A, Serhrouchni M, Hoerter J, Fischmeister R. Cardiovascular effects of *Urtica dioica* L. in isolated rat heart and aorta. Phytother Res 2002; 16(6):503-507.
- Prasana KK, Lilakanth N, Suvakanta D, Sutharson L, Bhagabat N. Hepatoprotective effect of the ethanolic extract of *Urtica parviflora* Roxb. in CCl₄ treated rats. International Journal of Pharmacology 2007; 3: 362-366.
- Kavalali G, Tuncel H, Goksel S, Hatemi HH. Hypoglycemic activity of *Urtica pilulifera* in streptozotocin-diabetic rats. J Ethnopharmacol 2003; 84:241-245.
- Farzami B, Ahmadvand D, Vardasbi S, Majin F.J, Khaghani S. Induction of insulin secretion by a component of *Urtica dioica* leave extract in perfused Islets of Langerhans and its vivo effects in normal and streptozotocin diabetic rats. J Ethnopharmacol 2003;89: 47-53.

12. Bnouham M, Merhfouf FZ, Ziyyat A, Mekhfi H, Aziz M, Legssyer A. Antihyperglycemic activity of the aqueous extract of *Urtica dioica*. *Fitoterapia* 2003; 74:677-681.
13. Aslan M, Deliorman Orhan D, Orhan N, Sezik E, Yesilada E. In vivo antidiabetic and antioxidant potential of *Helichrysum plicatum* ssp. *Plicatum capitulums* in streptozotocin-induced diabetic rats. *J Ethnopharmacol* 2007; 109: 54-59.
14. Harbone JB. 1984. *Phytochemical Methods. A guide to Modern Techniques of Plant Analysis*, second ed. Chapman and Hall, London, pp. 84-274.
15. Pan GY, Huang ZJ, Wang GJ, Fawcett JP, Liu XD, Zhao XC, Sun JG, Xie YY. The antihyperglycaemic activity of berberine arises from a decrease of glucose absorption. *Planta Med.* 2003; 69: 632–636.
16. Quanhong L, Caili F, Yukui R, Guanghui H, Tongyi C. Effects of protein-bound polysaccharide isolated from pumpkin on insulin in diabetic rats. *Plant Foods for Human Nutrition* 2005; 60:13-16.
17. Rao AV, Gurfinkel DM. The bioactivity of saponins: triterpenoid and steroidal glycosides. *Drug Metabol. Drug Int.* 2000; 17: 211-235.
18. Mukherjee PK, Maiti K, Mukherjee K, Houghton PJ. Leads from Indian medicinal plants with hypoglycemic potentials. *J Ethnopharmacol* 2006; 106:1-28.