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Evaluation of Antidiabetic Potential *of Ipomoea turpethum* R.Br. and *Ipomoea batata* L. (Convolvulaceae) in Alloxan Induced Diabetes in Rats: A Comparative Study.

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ABSTRACT

In order to explore the intra-genera variation for biological activity, the antidiabetic activity of *Ipomoea turpethum* and *Ipomoea batata* was carried out in alloxan induced diabetes in rats. Dried powdered material of both plants was defatted with petroleum ether 60-80 °C and cold macerated with hydro-ethanol for seven days. These extracts were screened for antidiabetic activity using alloxan induced diabetes in rats. Diabetes was induced by intra-peritoneal administration of alloxan monohydrate (120 mg/kg) on days 1 and 12 and blood glucose levels were estimated on 15th day. Hydro-ethanol extracts of both the plants at the dose of 500 mg/kg, oral, significantly lowered the blood glucose levels in diabetic rats treated for 7 days. The antidiabetic activity of these plants was insignificantly different from each other. This suggests that, different species from same genera may have similar chemical constituents and pharmacological activity.

Keywords: *Ipomoea turpethum*, *Ipomoea batata*, Diabetes, Hypoglycaemic, Comparative evaluation

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INTRODUCTION

Hitherto numerous oral allopathic drugs are available in the market for the treatment of diabetes and are associated with toxic effects [1]. However, treatment with traditional system of medicine was found to be beneficial with less or no toxic effects [2]. Therefore, scientific exploration of medicinal plants on the basis of their traditional claim has been encouraged.

Ipomoea turpethum (Convolvulaceae) is a large stout perennial twine vernacularly called 'Nishoth' or 'Nishottar'. The roots of *I. turpethum* are used against many indications including purgative, anthelmentic, hepatic stimulant, obesity, etc [3-4]. Interestingly, ayurvedic book Chikitsa Prabhakar recommends the use of *I. turpethum* in the treatment of diabetes [5], but there is no scientific data available for it. *Ipomoea batata* (Convolvulaceae) is a slender prostrate vine with tuberous roots, simple, alternate, ovate-cordate and angular or deeply lobed leaves [3-4]. In the traditional system of medicine, it is used in burning sensation, constipation, hyperdipsia, renal and vesical calculi, and diabetes [3, 6].

Furthermore, it was observed that most of the species in same genera have similar constituents and pharmacological activities. However, due to species, climatic and environmental differences; the quantity, constituents and pharmacological activity also changes [7]. Under certain climatic conditions, the non explored species may have better composition of chemical constituents and good pharmacological activity as compared to widely used allied species. *I. turpethum* is less employed and used rarely as compared to *I. batata*. It has many chemical constituents similar to *I. batata* and *I. aquatica* such as gentisic acid, p-coumaric acid, saponins, kaemferol, etc [7]. Based upon these evidences, it appears that both the plants may have antidiabetic activity and therefore, we have carried out comparative evaluation of the antidiabetic potential of *I. turpethum* and *I. batata* in alloxan induced diabetic rat.

MATERIALS AND METHODS

Plants

For the present study, roots of *I. turpethum* were collected in November from the fields of College of Agriculture, Panjabrao Deshmukh Agricultural University, Nagpur and rhizomes of *I. batata* were collected in November from the field 40 km from Nagpur and shade dried. Both the plants were identified and authenticated by Dr. (Mrs.) Alka Chaturvedi, Department of Botany, Rashtrasant Tukadoji Maharaj Nagpur University, Nagpur. Voucher specimens of *I. turpethum* and *I. batata* were deposited in the herbarium of Department of Botany as specimen No. 6558 and 9062 respectively.

Chemicals and instruments

Alloxan monohydrate was purchased from Sigma Chemicals (St. Louis, USA). All the solvents used were of analytical grade and purchased from Qualigens fine chemicals division, GlaxoSmithKline Ltd (Mumbai, India). Metformin hydrochloride was generously donated by M/s Zim Laboratories Ltd (Nagpur, India) as gift sample. Glucometer with test strips for estimation of blood glucose levels were procured from Abott Labs (Mumbai, India).

Extraction

Dried, coarsely powdered roots and rhizomes of both the plants were defatted with petroleum ether and cold macerated with hydro-ethanol (30:70) for seven days. The hydro-ethanol extracts obtained were concentrated in rotary vacuum evaporator, dried in vacuum oven and stored in desiccators (Yields: 3.63% w/w for *I. turpethum* and 4.03% w/w for *I. batata)*.

Animals

Male albino rats (Sprague-Dawley strain, weighing 200-250 g) from the animal house of the Department were used for the studies. All the animals were maintained under laboratory temperature (25±2 °C), humidity (45±5%) and 12 h light: 12 h dark cycle. They were allowed a standard pellet diet (Trimurti

Rodent Feeds, Nagpur, India) and tap water ad libitum. Institutional Animal Ethical Committee (IAEC), constituted under the guidelines of Committee for Purpose of Control and Supervision of Experimental Animals (CPCSEA), Ministry of Environment, Govt. of India, New Delhi approved all the animal experimental protocols.

Acute toxicity study: determination of LD50

The toxicological effects of both the plants were observed for gross behavioural, neurological and anatomical continuously for 2 h and then at 6 h intervals for 24 h as per guidelines-425 of Organization for Economic Cooperation and Development (OECD) [8].

Induction of diabetes and treatments

Diabetes was induced by intra peritoneal (i.p.) administration of alloxan monohydrate in saline (120 mg/kg) on days 1 and 12 [9]. The rats were given 2% glucose orally in order to prevent alloxan induced transient hypoglycaemia. The blood samples were collected from tail vein of rats on day 15 and blood glucose levels were estimated. Rats having blood glucose levels above 200 mg/dl were selected for further experiments [10] and divided in five groups of five rats each. Group I (normal untreated rats fed 2 ml of CMC, 0.5% w/v); group II (diabetic untreated rats fed 2 ml of CMC, 0.5%w/v); groups III and IV (diabetic rats treated with hydro-ethanol extract of *I. turpethum* (IT200; 200 mg/kg) and (IT500; 500 mg/kg) respectively), groups V and VI [diabetic rats treated with hydro-ethanol extract of *I. batata* (IB200; 200 mg/kg) and (IB500; 500 mg/kg) respectively),and group VII (diabetic rats treated with 500 mg/kg of metformin). All groups were treated once a day for 7 days by suspending each extract in distilled water with the help of CMC (0.5% w/v) and given orally using an intra-gastrictube

Collection and processing of blood samples for estimation of glucose levels

The blood samples, collected from tail vein, were analyzed by glucometer. The blood glucose levels were evaluated at regular time intervals at 0, 1, 3 and 5 h after single dose treatment (acute treatment). In the sub-acute treatment protocol, blood glucose levels were estimated 1 h after the treatment on days 3 and 7.

Statistical Analysis

All the data were analyzed by one-way analysis of variance (ANOVA) followed by Newman-Keul's *t*-test for multiple comparisons. $p < 0.05$ was considered significant.

RESULTS AND DISCUSSION

Acute toxicity study: determination of LD50

The oral acute toxicity for hydro-ethanol extracts of *I. turpethum* and *I. batata* were carried out as per OECD guidelines-425 and the median lethal dose (LD_{50}) was found to be > 2000 mg/kg for both the plants. In addition, previously Gopi and his co-workers reported no mortality at single oral dose of 4000 mg/kg of *I. Turpethum* [10].

Induction of diabetes and treatments

Acute effect of treatment

Intra-peritoneal (i.p.) administration of alloxan monohydrate (120 mg/kg) on day 1 and 12 [11] significantly elevated the blood glucose levels (BGL, > 200 mg/dl), which on treatment with hydro-ethanol extract of *I. turpethum*, *I. batata* and metformin were compared with untreated diabetic rats. Except meformin, hydro-ethanol extract of *I. turpethum* (IT500; 500 mg/kg) and *I. batata* (IB500; 500 mg/kg) did not show significant decrease in BGL during the first 3 h of treatment, whereas after 5 h IT500, IB500 and metformin lowered the BGL significantly by 41%, 38% and 43% respectively as compared to control group. The lower doses of hydro-ethanol extracts of *I. turpethum* (IT200; 200 mg/kg) and *I. batata* (IB200; 200 mg/kg)

failed to lower the BGL at any time points. Comparing the activities of IT500 and IB500, it was found that both extracts showed insignificant difference in lowering the BGL.

Sub-acute effect of treatment

In order to study the long term effects of extracts, the treatment was given for 7 days and BGL were measured on days 1, 3 and 7. Following IT500, IB500 and metformin treatments the BGL were decreased on day 1 by 46%, 44% and 49%, on day 3 by 53%, 54% and 60% whereas on day 7 by 56%, 55% and 64% respectively.

Although, the traditional therapy claims use of *I. turpethum* in the treatment of diabetes [5], no scientific data substantiate this claim. The experimental observations in the present study establish evidences for the efficacy of *I. turpethum* in the treatment of diabetes. Meformin, hydro-ethanol extract of *I. turpethum* (IT500; 500 mg/kg) and *I. batata* (IB500; 500 mg/kg) showed significant decrease in BGL after 5 h of oral administration when compared with untreated diabetic rats. However, during the first 3 h of treatment, only metformin could lower the BGL significantly. Furthermore, phytochemical investigation of *I. turpethum* confirmed the presence of flavonoids, tannins, polyphenols and many of the chemical constituents which are common to other Ipomoea species having antidiabetic property [7]. Over 150 plant extracts and some of their active principles including flavonoids such as kaempferol are known to be used for the treatment of diabetes [12-14]. Moreover, tannin containing drugs demonstrated antidiabetic activity [15-17].

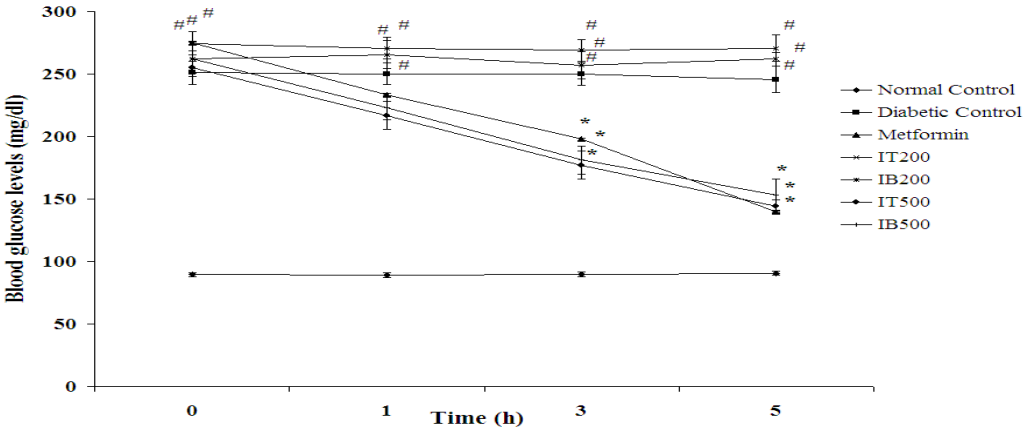

Figure 1: Effect of acute oral treatment of hydro-ethanol extracts of *Ipomoea turpethum* R.Br. and *Ipomoea batata* L. on alloxan induced diabetes in rats. Results are expressed as mean± SEM (n=5). # p<0.001 Vs normal control, * p<0.01 Vs diabetic control.

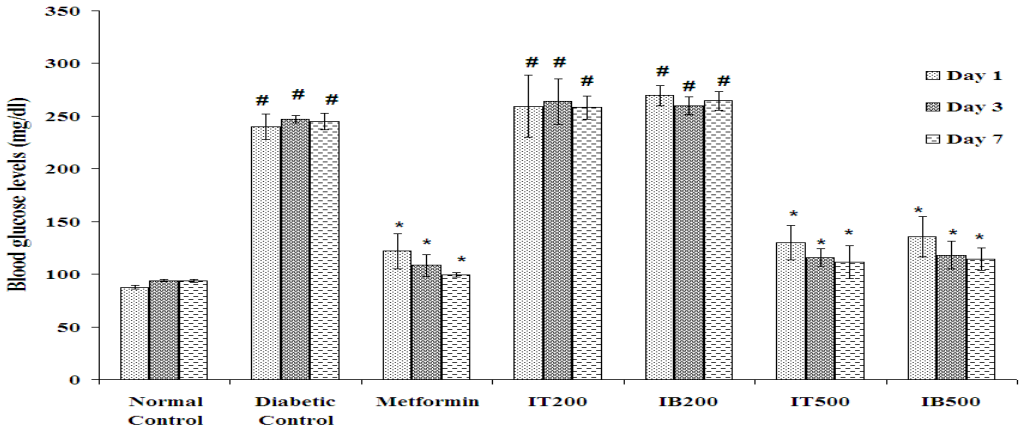

Figure 2: Effect of chronic oral treatment of hydro-ethanol extracts of *Ipomoea turpethum* R.Br. and *Ipomoea batata* L. on alloxan induced diabetes in rats. Results are expressed as mean±SEM (n=5). # p<0.001 Vs normal control, * p<0.01 Vs diabetic control

Kusano and his co-workers have shown that, though *I. batata* improves glucose tolerance and decreases BGL, it increases the blood insulin levels in streptozotocin diabetic rats [6,8]. Another interesting fact about *I. batata* is that, it decreases the insulin resistance with no effect on body weight, or secretion and clearance of insulin [9, 18]. These studies suggest that *I. batata* has unique advantageous effect in the treatment of both insulin deficient as well as insulin resistance diabetes. The beneficial effects of antioxidants are well known in diseases like cancer and diabetes [19]. Coincidently the body defence mechanism has an army of some antioxidant enzymes such as superoxide dismutase (SOD) and catalase. Gopi and his co-workers reported the increase in activities of the SOD and catalase following administration of hydro-alcoholic root extract of *I. Turpethum* [10]. On the basis of the above reports, it is possible that the presence of flavonoids and tannins are responsible for the observed antidiabetic activity of *I. turpethum*.

This study substantiates the traditional antidiabetic claim of *I. turpethum*. As the antidiabetic activity of IT500 and IB500 is insignificantly different from each other, it reveals that, the different species from the same genera having similar chemical constituents can possess the identical pharmacological activity. This activity can be attributed to a particular class of chemical constituents.

CONCLUSION

The results of this study indicated that, *I. turpethum* possesses antidiabetic activity and found to be at par with *I. batata* and metformin in alloxan induced diabetic rats. This might be due to the presence of flavonoids and tannins in both the species.

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