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Evaluation of anti-inflammatory activity of *Trichosanthes dioica* R. Leaves

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Abstract
Trichosanthes dioica R. (*T. dioica*) is an easily available common plant. The plant belongs to family Cucurbitaceae which has given us many important medicinal plants like *Momordica charantia, Citrullus colocynthis* etc. from which important pharmacological activities and markers like charantin and Cucurbitacin have been reported and isolated. In the present study *T. dioica* was evaluated for anti-inflammatory activity using carrageenan induced acute inflammation model in rats. It was concluded that leaves of *T. dioica* possess moderate anti-inflammatory activity.

Keywords: *Trichosanthes dioica*, carrageenan, anti-inflammatory activity

Introduction
Trichosanthes dioica R. is an important medicinal herb. In Charak Samhita, leaves and fruits used for treatment of alcoholism, jaundice, oedema and alopecia.[1] Over 20 species of *Trichosanthes* are recorded in Asia of which two namely *T. dioica* and *T. anguina* are cultivated as vegetable. [2] *Trichosanthes dioica* (Pointed gourd) is known by the name of *parwal*, *palwal*, *parmal, patol, potala* in different parts of India and Bangladesh and used as antipyretic, diuretic, cardiotonic and laxative [3]. The fruit and leaves is the edible part of the plant which is cooked in various ways either alone or in combination with other vegetables or meats [4]. Juice of leaves of *T. dioica* is used as tonfmic, febrifuge and in subacute cases of enlargement of liver and spleen. [5] The various chemical constituents present in *T. dioica* are vitamin A, vitamin C, tannins, saponins. [6] The leaves of *T. dioica* are used in oedema ethnopharmacologically (Shaarangadhara Samhita), but no scientific data for both activities is available yet. Therefore it was thought to investigate anti-inflammatory potential of the leaves of *T. dioica* using carrageenan induced acute inflammation model in rats.

Materials and Methods
Collection and authentication of plant material
Leaves of *T. dioica* were collected from local area of Lucknow (U.P.) India, during the month of October and authenticated by Division of Taxonomy, National Botanical Research Institute (NBRI), Lucknow, India and a voucher specimen was deposited for future references (Ref. No. NBRI/CIF/128/2010).

Animals
Healthy male Wistar albino rats each weighing 100-120g were used for study. The rats were housed in polypropylene cages and maintained under standard conditions (12 h light and dark cycles, at 25 $\pm 3^0$C and 35-60% humidity). Standard pelletized feed and tap water was provided *ad libitum.*

Ethical approval
The experiments were performed as per protocols set by CPCSEA which was duly approved by Institutional Animal Ethical Committee (Approval No. BBDNITM/IAEC/23/2010).

Acute oral toxicity
Wistar albino rats, fasting, for 24 h were administered, ethanolic and aqueous extracts of *T. dioica* leaves at 2000 mg/kg, p.o. The animal were observed for 24 h. the animal survived and therefore 4 more animals were dosed at the same dose i.e. 2000 mg/kg, p.o. and were observed for 24 h. [7]

Experimental procedure for anti-inflammatory activity

Carrageenan induced rat paw edema

Four groups of albino rats having five animals in each were formed. Group-A Treated orally with vehicle (3ml/kg), Group-B Diclofenac (12.5 mg/kg), Group-C Aqueous extract, Group-D Ethanol extract of leaves suspended in the vehicle, 60 min prior to an injection of 0.1 ml 1% carrageenan into the plantar tissue of the right hind paw. Paw volume of each animal of all groups was measured by plethysmography at 0, 1, 2 and 3 h after carrageenan injection.

Edema was assessed in terms of volume of mercury displaced by the paw before and at 1, 2, & 3 hours after induction of inflammation [12, 13]

Inhibition (%) = 100[1- (a-x/b-y)]

Where

a = mean paw volume of treated animals after carrageenan injection

x = mean paw volume of treated animals before carrageenan injection

b = mean paw volume of control animals after carrageenan injection

y = mean paw volume of control animals before carrageenan injection

Statistical analysis

The results were expressed as mean ± SEM and statistically analyzed by ANOVA followed by Dunnett's test, with level of significance set at $p<0.05$ and $p<0.01$.

Results

Acute oral toxicity

Wistar albino rats, fasting, for 24 h were administered, ethanolic and aqueous extracts of *T. dioica* at 2000 mg/kg, p.o. The animal was observed for 24 h. the animal survived and therefore 4 more animals were dosed at the same dose i.e. 2000 mg/kg, p.o. and were observed for 24 h. All five animals survived. Therefore 2000 mg/kg dose was considered safe and 1/10th of the dose was selected for further evaluation.

Anti-inflammatory effect

Table 1: Effect of extracts on paw edema induced by carrageenan in rats

Groups	Dose (mg/kg)	Mean paw volume ± SEM (ml) and % Inhibition			
		Before Carrageenan	Time after Carrageenan injection		
		0h	1h	2h	3h
A Control	-	0.32±0.007	0.51±0.01	0.49±0.004	0.50±0.01
B Standard	12.5	0.34±0.007	0.37±0.007** (84.21%)	0.36±0.01** (88.23%)	0.36±0.01** (88.88%)
C Ethanolic extract	200	0.34±0.01	0.47±0.01* (31.57%)	0.45±0.01** (35.29%)	0.44±0.01** (44.44%)
D Aqueous extract	200	0.31±0.007	0.47±0.01* (15.78%)	0.43±0.008** (29.41%)	0.42±0.01** (38.88%)

Values are expressed as mean ± SEM, n=5, *P<0.05, **P<0.01 considered as significant (compared with control group) using one way ANOVA followed by Dunnett's test

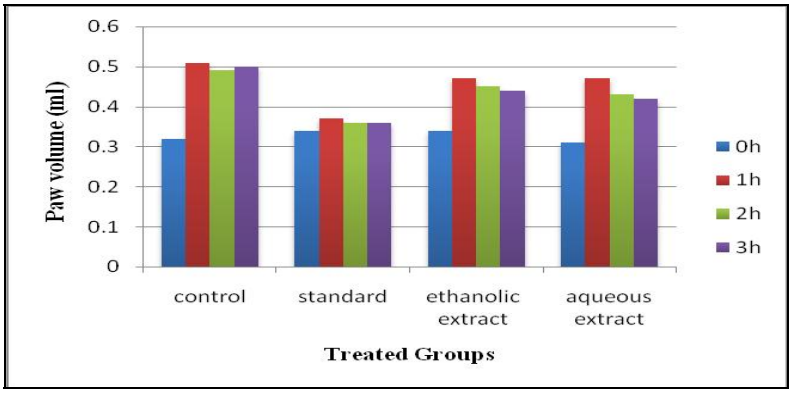

Fig 1: Graph showing the effects of extracts on paw volume of albino rats

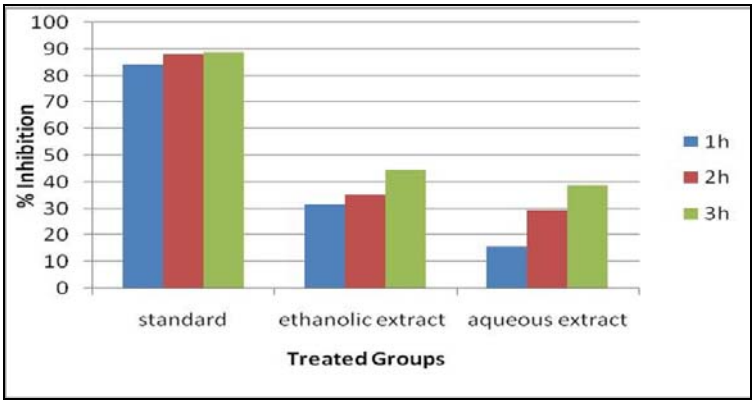

Fig 2: Graph showing % inhibition of paw edema in treated groups

Discussion

In carrageenan-induced acute inflammation model paw volume of each animal of all groups was measured by plethysmograph method, at 0 h (before) & 1, 2, 3 h after carrageenan injection (Table 1). In the duration of first hour aqueous extract (200 mg/kg) showed the mild anti-infammatory activity i.e. 15.78% inhibition of inflammation, but ethanolic extract (200 mg/kg) showed 31.57% inhibition of inflammation. This initial potential of ethanolic extract indicated the more potency of it in comparison of aqueous extract (200 mg/kg). In the duration of second hour anti-inflammatory activity of aqueous extract increased to 29.41%, ethanolic extract 35.29% & standard (Diclofenac) 88.23%. In the duration of third hour anti-inflammatory activity of aqueous extract increased to 38.88%, ethanolic extract 44.44% & diclofenac (12.5 mg/kg) 88.88%. From the graph shown in Figure 1 & 2, it can be suggested that both the extracts have exhibited the inhibitory effect but lesser than standard.

Thus in the duration of 3 hours, the ethanolic extract (200 mg/kg) showed better inhibition of rat paw edema than aqueous extract. It was nearly half to the standard drug diclofenac (12.5 mg/kg, p.o.).

So it can be suggested that both the exracts have anti-inflammatory effect but ethanolic extract have much better effect (44.44% inhibition of inflammation) than effect of aqueous extract (38.88% inhibition of inflammation) at the end of 3h. Thus the edema suppressant effects of both the extracts were found to be moderate at the end of 3 hours & significant (p<0.01) as compared to control.

Conclusion

From the pharmacological study for anti-inflammatory activity of *T. dioica* leaves it can be concluded that both the ethanolic & aqueous extracts of leaves at 200 mg/kg dose posses significant anti-inflammatory activity than control. Although the protections provided by ethanolic & aqueous extracts were less as compare to standard still both has a potential to be used as moderate anti-inflammatory agent. It can also be concluded that ethanolic extract at 200 mg/kg dose posses superior anti-inflammatory activity than aqueous extract of *T. dioica* at the similar dose level. The study in the direction of elucidating the mechanism of anti-inflammatory activity needs to be conducted.

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