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RESEARCH ARTICLE

IN-VIVO ANTI-INFLAMMATORY ACTIVITY OF METHANOLIC EXTRACT OF *WATTAKAKA VOLUBILIS* (ASCLEPIADACEAE)

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ABSTRACT

The present work evaluated the anti-inflammatory activity of methanolic extract of *Wattakaka volubilis* against the carrageenan induced rat paw oedema in Wister Albino rats at dose dependent manner (150 and 300mg/kgbody weight). Our results showed that the methanolic extract of *W. volubilis* gives a strongest anti-inflammatory activity (71.7%) werecompare to the standard drug as indomethacin.

INTRODUCTION

Wattakaka volubilis belong to the family Asclepiadaceae it is used as cure to various ailments since ancient times (Pullaiah, 2002). *Wattakaka volubilis* is a tall woody climber, young branches green, slender, smooth, leaves opposite. It isearlier reported on thisplantareanti-inflammatory, analgesic and anti-lipid peroxidative (Divya *et al.,* 2009), The plant parts has been traditionally used for medicinal purposes. This plant juice used tosternutatory and leaves are employed in application for boils and abscesses (The Useful medicinal plants of India, 1992). Various phytochemical constituents like steroids, steroidal glycosides, sugars, triterprnoids, flavonoids, phenolic compounds and some alkaloids are found to be present in the plant (Anonymous, 2003). *in vitro* anti-leishmanial and antitumour, hepatotoxicity, prevention of proteolysis in rat lens (Molisha *et al.,*2009; Biju *et al.,*2007).

MATERIALS AND METHODS

The whole plant of *W.volubilis* was collected from Velliangiri hills, Coimbatore district Tamilnadu (India) and authenticated by a taxonomist, M. Murugaesan, SACON, Coimbatore. The plant material was dried in shade and then powdered using pulveriser and passed through 100 mesh sieve.

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About 100 g of dried plant powder was defatted with petroleum ether used for this study re-extracted with methanol. This extract after evaporation of methanol, the filtered residue was stored at 4 ^{0}C in refrigerator.

Maintaining the animals

The extract was subjected for acute and sub-acute toxicity studies using animal model and LD_{50} doses were determined for the pharmacological activity. The methanolic extract was used as an emulsion in 5% suspension with gum acacia and administered orally at the dose of 150and 300mg/kg b.wt. The animals were grouped in cage in an air conditioned room at the temperature of 22±1ºC with 12 hour light and dark cycle. The animals were maintained with pellet diet and water *ad libitum*. They were further segregated into various groups. This experiment was performed according to ethical guidelines for the investigation of experimental pain in conscious animals (659/02/a/CPCSEA).

Carrageenan-induced paw oedema in albino rats (Winter and Poster, 1957)

Rats were divided into 4 groups comprising five Rats in each group. In all groups acute inflammation was produced by sub plantar injection of 0.1ml freshly prepared 1% suspension of carrageenan in normal saline in the right hind paw of the rats and paw volume was measured plethysommetrically at 0 to 180mins after carrageenan injection. All the animals were premedicated with indomethacin (10mg/kg b.wt.) orally two hour before infection.

Table 1. *In vivo* anti-inflammatory activity of *W. volubilis* methanolic extract of Carrageenan induced hind paw oedema in rats

Treatment	Oedema volume (ml)					% Inhibition after 180 min
	Dose mg/kg	0 min	60 min	120 min	180 min	
Group I	Normal saline	24.13±1.83	42.16±1.08	31.13±1.65	131.84±1.84	_
Group II	150µg/kg	19.63±1.93	57.16±1.29	67.23±1.52*	59.41±1.2*	55
Group III	300µg/kg	21.04±1.22	61.84±1.37**	39.26±1.93**	37.21±1.39***	71.7
Group IV	10mg/kg	29.13±1.62	42.66±1.39**	36.12±1.75	25.73±1.21***	80.6

Each Value is SEM ± 5 individual observations * P <0.05 ; ** P<0.01 ;* ** P<0.001 Compared paw oedema induced control vs drug treated rats

Mean increase in paw volume was measured and percentage was calculated for all the extracts. These extracts were subjected for acute toxicity studies and 1/10th of the LD_{50} dose was selected for pharmacological activity. Percentage inhibition of paw volume was calculated by the following formula

$$\% \text{ inhibition} = \frac{Vc - Vt}{Vc} \times 100$$

Where

Vt- means increase in paw volume in rats treated with test compounds
Vc- means increase in paw volume in control group of rats.

Statistical analysis

The mean paw volume was expressed in terms of mean ± SEM and evaluated for statistical significance by ANOVA followed by Dunnett's test, P<0.05 was considered by statistically significant.

RESULTS AND DISCUSSION

In the present study anti-inflammatory activity of*W. Volubilis* methanolic extract (150mg/kg and 300mg/kg, b.wt.) significantly (P < 0.01) reduced the mean paw edema volume at 3 h after carrageenan injection. The *W. volubilis* methanolic extracts treated groups exhibited improved anti-inflammatory activity with the percent inhibition of paw edema of 55and 71.7mg/ Kg b.wt. respectively, as compared with the control group. However, the standard drug, indomethacin (10mg/kg b.wt.,) showed highly significant (p<0.001) anti-inflammatory activity with the percent inhibition of 80.6 as shown in Table 1. Carrageenan injection into the rat paw provokes a local, acute inflammatory reaction that is a suitable criterion for evaluation of anti-inflammatory agents (Winter *et al.,* 1962).

The time course of oedema development in Carrageenan induced model in rats is generally represented by a biphasic curve (Vinegar *et al.,* 1969).Non-steroidal anti-inflammatory drugs (NSAID) such as indomethacin used in this study are known to inhibit cyclooxygenase enzymes I and II which are implicated in the production of inflammation- mediating agent prostaglandin E2 (PGE2) from arachidonic acid (Dhara *et al.,* 2000, Wu, 2003, Moody *et al.,* 2006).Nabiland Leila. (2016) reported that anti-inflammatory activity *in vivo* by the paw edema assay induced by carrageenan showed that oral administration of MeOHE at a dose of 200 mg/kg in rats treated with carrageenan causes a significant decrease of inflammation compared with the control group and which is slightly greater than the effect of diclofenac that was used as a positive control.

The analysis of C-reactive protein shows the absence of this protein in the plasma of rats treated with MeOHE of the plant. As regards the analgesic activity of the MeOHE has a very significant reduction in numbers of abdominal writhes at a dose of 400 mg/kg bw, these results are very similar to those obtained in the group treated with paracetamol.It can conclude from present study that *W. volubilis* methanolic extract have been used for development of standardized plant herbal therapeutic formulation for anti inflamatory conditions.

Group I: Control rats given normal saline orally by using an IntraGastric Catheter tube (IGC).

Group II: Rats given methanolic*W. volubilis* extract at the dose of 150 mg/ Kg b.wt. by IGC.

Group III: Rats given methanolic *W. volubilis* extract at the dose of 300 mg/ Kg b.wt.by IGC.

Group IV: Rats given Indomethacin at the dose of 10 mg/ Kg b.wt.by IGC.

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