
Antiinflammatory Activity of the Volatile Oil of *Toddalia Asiatica*

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Oil of *Toddalia asiatica* leaves obtained by steam distillation was given orally to study its effects on the exudative and proliferative phases of the inflammatory reactions, using the technique of carrageenin induced paw edema and cotton pellets in male albino rats. The antiinflammatory activity was compared with ketorolac tromethamine. In carrageenin-induced paw edema, 0.8 ml/kg of the volatile oil showed antiinflammatory activity comparable to that of ketorolac tromethamine. The oil was also found to be effective in cotton pellet granuloma studies.

T. ASIATICA has been used as a household remedy for local application in inflammatory conditions and other painful conditions including sprains (1). Patnaik et al. (1986) reported the antiinflammatory activity of alcoholic extract of *T. asiatica* (2). From the alcoholic extract of *T. asiatica* a new benzoquinoline together with six alkaloids and six coumarins were isolated (3). The unripened fruits and roots are rubbed with oil to prepare a stimulant liniment for use in rheumatism. *T. asiatica* leaves distilled yield a yellowish green essential oil of sharp aromatic odour like that of citron containing citronella aldehyde. The chief constituent is a camphor like body with a melting point of 96.5°-97°C. Citronellal and linalol are also present in it. (4) We investigated the effect of volatile oil of *T. asiatica* leaves in different phases of inflammation.

Materials and Methods :

Volatile oil was extracted from the fresh leaves of *T. asiatica* by steam distillation. This oil was used after emulsifying in gum acacia (5%) for oral administration. Male albino rats weighing between 150 to 200 g bred in King Institute Guindy, Madras were selected for the studies. Antiinflammatory activity

was studied by carrageenin-induced rat hind paw edemas and cotton pellet granuloma methods after conducting acute toxicity studies.

Acute Toxicity studies:

Animals were divided into 5 groups consisting of 5 animals, in each group. One group served as control. For remaining four groups, oil was administered orally in different doses of 0.2, 0.4, 0.8 and 1.6 ml/kg.

Following the drug administration during first 2 hr., animals were observed for gross behavioural changes (behavioural, neurological and autonomic responses). Animals observed once in half an hour for next 4 hr. and then once in 25 hr. to find out the percentage of mortality.

Carrageenin-induced edema :

The rats were divided into 5 groups, each group consisting of 10 animals. One group served as negative control (received 5% Gum acacia soln. 5 ml/kg), the second group served as positive control (received ketorolac tromethamine 10 mg/kg) while the other groups received essential oil in different doses of 0.2, 0.4 and 0.8 ml/kg orally.

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Edema was produced by the method described by Winter *et al.* (5). The paw volume was measured 0 hr. and 3 hr., after the injection of carrageenin. The apparatus used for the measurement of rat paw volume was that of Buttle *et al.* (1966) modified by Singh and Ghosh (6). This method, followed here in our laboratory is able to detect, a minimal change of paw volume of + 0.02 ml. Drug pre-treatment was given 1 hr. before the injection of carrageenin. The percentage inhibition of edema was calculated.

Cotton Pellet Granuloma :

Albino rats were divided into 4 groups, each group consisting of 8 animals. One group served as negative control (received 5% gum acacia 5 ml/kg body wt.) the second group served as positive control (received ketorolac tromethamine 10 mg/kg body wt.) and the remaining two groups received the volatile oil of *T. asiatica* leaves (0.4 and 0.8 ml/kg body wt. respectively). After shaving off the fur on the back, the rats were anesthetized with pentobarbitone (30 mg/kg). Through a single middle incision on the dorsal surface, sterilised preweighed cotton pellets were implanted in both axillae and groins according to the methods of D'Arcy *et al.* (1960) with slight modification. The drugs were administered daily usually for 10 days (0 to 9 days). On the 10th day the pellets were dissected out and dried at 60°C, and the dry weight was obtained.

Granuloma weight was obtained by deducting the weight of the cotton pellets on '0' day (i.e., before start of the experiment) from the weight of the cotton pellet on '9th' day (i.e., at the end of the experiment)

RESULTS

Acute Toxicity Study

The essential oil of *T. asiatica* leaves were found to be safe upto a maximum dose of 1.6 ml/kg. There was no mortality and no changes in behavioural, neurological and autonomic responses were observed.

Table I shows the effect of drug treatments on carrageenin-induced edema. The results were analysed by analysis of variance. (8) Edema suppressant effect of 0.8 ml/kg dose of the oil was 44.01 ± 10.16 which was near equivalent to that of 10 ml/kg dose of ketorolac tromethamine is 51.6 ± 10.47 . The edema suppressant effect was significant (at $p < 0.05$) in all dose levels except 0.2 ml/kg when compared to control. Though the oil showed dose response inhibition of inflammation, it was not significant among all test dose levels.

Table II shows the effect of drug treatment on the mean weights of cotton pellets. The oil at both the dose levels. (0.4 ml and 0.8 ml/kg) inhibited the granuloma tissue formation showing significant dose-proportionate inhibitory effect on the granuloma weight. The inhibitory effect of oil at the dose of 0.8 ml/kg body weight was found to be similar to that of ketorolac tromethamine 10 ml/kg.

DISCUSSION

The oil of *T. asiatica* leaves seems to be very safe as it did not show any toxicity in acute toxicity studies. The 10 day administration of the oil also did not show any toxic manifestations.

Carrageenin-induced paw edema was taken as a prototype of exudative phase of inflammation. The development of edema has been described as biphasic (9). The initial phase is attributable to the release of histamine, serotonin and kinin in the first hour after injection of carrageenin. A more pronounced second phase is related to the release of prostaglandins like substances in 2-3 hours. The significant anti-inflammatory effect of oil of *T. asiatica* leaves, like alcoholic extract of root bark seems to be related to its histamine, kinin and prostaglandin inhibitory activity (10 & 11).

In the cotton pellet granuloma model, inflammation and granuloma develops during a period of several days. This model is the indication for the

Table I: Effect of volatile oil of *T. asiatica* on carrageenin induced paw edema

Method : Carrageenin rat paw edema
 Animals : Albino rats
 Weight : 150-200g
 Vehicle : 5% Gum acacia.
 Route of administration — Oral
 Number of animals — 10 in each group.

Sl. No.	Drug	Dose ml/kg body weight	Increase in paw volume after 3 hrs mean \pm SEM	% Decrease in paw volume mean \pm SEM	P Values	
					Vs Control	Vs Different doses
1.	Control 5% Gum acacia	5 ml/kg of body weight	0.334 \pm 0.032 \pm			
2.	Standard Ketorolac tromethamine	10 mg/kg	0.162 \pm 0.030 \pm	51.6 \pm 10.47	P<0.05*	
3.	Oil	0.2 ml/kg body weight	0.226 \pm 0.051	32.34 \pm 9.87	p<0.05*	NS
4.	Oil	0.4 ml/kg body weight	0.208 \pm 0.023	37.72 \pm 9.12	P<0.05*	NS
5.	Oil	0.8 ml/kg body weight	0.187 \pm 0.016	44.01 \pm 10.16	P<0.05*	NS

* = significant difference

NS = Non significant

proliferative phase of inflammation. Inflammation involves proliferation of macrophages, neutrophils and fibroblasts, which are basic sources for granuloma formation. Therefore, decrease in granuloma weight indicates the suppression of proliferation phase which was effectively inhibited by *T. asiatica* leaf oil as indicated by our study.

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Table 2

Sl. No.	Drugs	Dose	Granulation weight in mg mean \pm SEM	% in granulation weight mean \pm SEM	P Values	
					Vs Control	Vs Different doses
1.	Control 5% Gum acacia	5 ml/kg of body weight	309 \pm 6.522			
2.	Ketorolac tromethamine	10 mg/kg	167.18 \pm 2.8	45.9 \pm 2.1	P<0.05*	
3.	Oil	0.4 ml/kg	202.30 \pm 4.52	34.53 \pm 3.3	P<0.05*	
4.	Oil	0.8 ml/kg body weight	0.187 \pm 0.0161	44.01 \pm 10.16	P<0.05*	P<0.05

* = Significant difference

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