Ancient science of life Vol No. 18(2), October 1998 pages 120 - 126

ANTI – INFLAMMATORY AND ANALGESIC PROPERTIES OF THE LEAVES OF TAMARINDUS INDICUS

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Received: 25 June, 1997 **Accepted: 1 August, 1998**

ABSTRACT: *The ethanolic extract of the leaves of Tamarindusindicus was investigated for its anti inflammatory and analgesic actions. The anti inflammatory activity was studied using carrageenin induced hind paw edema in rats and analgesic activity using chemical writhing test and tail clip method in mice and tail flick method yin rats. Test drug exhibited significant dose dependent anti inflammatory and analgesic and analgesic actions and found to be not superior to that of acetyl acid.*

INTRODUCTION

Different parts of *Tamarindus indicus* have been in use as herbal remedies for various ailments in Ayurveda and sidha. Poultices of leaves are recommended as applications to inflammatory swellings to relieve pain. A thick boiled syrup of pulp and leaves is used to heal swellings. Decoction of leaves is used as gargle in sore throat and for washing ulcers. The leaf extract has been reported to possess significant anti inflammatory as well as anti microbial properties. In the present study efforts were made to assess its anti inflammatory and analgesic activity in selected experimental animals and to compare these effects with known standard drugs.

MATERIALS AND METHODS

Solvent extraction of the dried leaves was done using 50% ethanol as per method by Rosenthaler (1930) and was dried to remove the solvent. A solution of the dried extract is distilled water was used for study. Animals such as mice and albino rats and equipments like modified (1930) and was dried to remove the solvent. A Solution of the dried extract is distilled water was used for study. Animals such as mice and albino rats and equipments like modified plethysmometer and anlgesiometer were used for the studies.

1. Anti inflammatory activity: Anti inflammatory activity was studied by the method described by winter et al (1960) using modified plethysmometer. (Hardayal Singh and Ghosh 1968).

Overnight fasted albino rats weighing 150-250g were divided into five groups of six animals. At zero hour the animals were given the following drugs orally.

Group I, Group II and Group III received the test drug in the dose of 100mg, 200mg and 500 mg per kg body weight respectively, while group IV received Acetyl salicytic acid 300 mg/kg body weight as standard and Group V received distilled water as control.

After 1 hour o.1 ml of 1% carrageenin was injected subcutaneously into the left hand paw of each animal and the initial reading (IR) noted. Three hours later the paw volume was again measured as the final reading (FR). From these two values the percentage inhibition of oedema was calculated using the formal.

$$\text{Percentage inhibition of oedema} = \frac{(C-D)}{C} \times 100$$

Where C is the mean difference in paw volume of the treated group. Food and water were restricted to the animals till the end of the study. The results were tabulated.

II. Analgesic activity

This was assessed by the following three tests.

1. Chemical writhing test in mice

Writhing syndrome was induced on albino mice (20-30g) by intraperitoneal injection of 3% acetic acid in a dose of 300 mg/kg body weight as per the method of witkin et al (1961). The syndrome was characterized by intermittent contractions of abdominal muscles with extension of hind limbs and twisting of trunks.

The 50% ethanol extract was administered orally in doses of 100mg, 200mg, and 500mg/kg body weight to groups of mice 30 minutes prior to the acetic acid injection for each dose level 6 mice were used. Another group of 6 mice were taken as standard and to them acetyl salicytic acid in a dose of 300 mg/kg body weight as standard and Group V received distilled water as control. group received the stretching episodes in each mouse was recorded for 30 minutes. Effect of drug was calculated by the percentage of inhibition of the stretching episodes over that of the control groups

2. Tail-clip method in mice

Method of binachi and David (1960) was followed. Albino mice of either sex (200-40g) were used for the study. A bull dog clip was applied gently to the base of the tail of the mouse and reaction time in seconds from the moment the clip was applied to the moment when the animal that did not respond within 5 seconds were discarded.

Such animals were then divided into 5 groups each containing 10 mice. They were fasted overnight After 20,60 and 90 minutes of drug administration, the tail clip was applied again and the reaction time was noted as above. The effect of the drug was calculated as the percentage of protection from removing the tail clip.

3. Tail flick method in rats.

This was studies as per the method of Gujral and Khanna (1957), the pain threshold induced by a hot nichrome analgesiometer was measured in rats by nothing the reaction time. Albino rats if nothing the reaction time. Albino rats of either sex (150-200g) in groups of eight was used for the study. The temperature of the wire was regulated and the normal reaction time for individual rat was measured prior to drugs by keeping each rat in a rat holder. Normal reaction time was the time taken by the animal for a sudden flick of tail when radiant heat is applied over the tail. The criteria for the end point was the failure of the animals to remove the tail clip.

The 50 % ethanol extract was given orally in doses of 100mg, 200 mg, and 500 mg/kg body weight to each group of rats. To another taken as standard was given acetyl salicylic acid orally in dose of 300mg/kg body weight. The control groups received distilled water. Pain threshold was measured by noting the reaction time at 30, 60,120,180 and 240 minutes after drug treatment. Percentages of analgesic activity was calculated form the control taken prior to the drug or vehicle in each group.

RESULTS

Table I shows that 50% ethanol extract of T. indicus in all the three doses provided significant protection against acute inflammation induced by carrageenin ($p< 0.05$). At 100mg/kg body weight, the test drug produced anti inflammatory effect of 44.82% where as at 200 mg and 500mg/kg doses the effects produced were 48.27% 51.73% respectively. But the standard drug acetyl salicylic acid at 300 mg/kg body weight showed 62.07% inhibition of inflammation ($p<0.01$)

The data shown in table 2 shows that the extract is capable of producing a dose dependent analgesic effect. The ethanol extract of T. indicus produced 47.38% inhibition of writhing syndrome ($p< 0.01$) at a dose of 200mg/lg body weight and 58% inhibition was obtained ($p<0.001$) at a dose of 500 mg/kg bodyweight. However salicylic acid, the standard drug showed 91.3% inhibition ($P<0.001$)

Table 3 shows that by tail clip test, maximum analgesic effect was shown at 500 mg/kg body weight within 30 minutes of administration ($P<0.05$), where as with 300 mg/kg dose of acetyl salicylic acid three was more protective effect ($P<0.01$). By tail flick test, the extract showed significant analgesia in various doses, but there was no dose dependent response. There was significant increase in reaction time which was evident after 30 minutes of drug administration and the analgesia lasted for about 180 minutes. But here also the potency was less compared to 300 mg/ kg body weight of acetyl salicylic acid (Table 4).

DISCUSSION

From the above results it could be seen that the ethanol extract of leaves of T. indicus possessed significant anti inflammatory effect as shown in acute inflammatory process. ($P<0.05$). However the effect was comparatively less when compared to the standard drug acetyl salicylic acid.

Analgesic activity tests showed that the extract possessed significant activity against mechanical, chemical and thermal pain stimuli. However the analgesic effect was not sustained. Moreover the extent and duration of acetyl salicylic acid, the standard drug used for comparative evaluation. The present studies corroborate with earlier reports (Vijayakumar et al 1993) but further studies are necessary to find our the exact mode of action.

ACKNOWLEDGEMENT

We are thankful to the staff of Dept of pharmacology, Medical college, Thiruvanathapuram and the CDRS Unit for their assistance.

Table -1
The Effect of Ethanol extract of T. indicus on acute inflammation

Drug	Dose (mg/ Kg b.w)	n	Mean increase in paw volume after 3 hrs in ml (Mean ± SE)	Percentage of anti-inflammatory activity	P value
Control (Distill water)	-	6	0.4833 ± 0.08	-	-
Standard (acetyl salicylic acid)	300	6	0.1833 ± 0.05	62.07	<0.01
Ethanolic extract	100	6	0.2667 ± 0.12	44.82	<0.1

Ethanolic extract	200	6	0.25 ± 0.05	48.27	<0.05
Ethanolic extract	500	6	0.2333 ± 0.05	51.73	<0.05

Table -II
The Effect of Ethanol extract of T. indicus on acetic acid induced writhing syndrome in albino mice

Drug	Dose (mg/ Kg b.w)	n	Mean increase in paw volume after 3 hrs in ml (Mean ± SE)	Percentage of anti-inflammatory activity	P value
Control (Distill water)	-	6	11.5±0.85	-	-
Standard (acetyl salicylic acid)	300	6	1.00±0.48	91.30	<0.001
	100	6	7.67±0.19	33.30	<.05
	200	6	6.00±1.16	47.83	<.01
Ethanolic extract	500	6	4.83±1.17	58	<.001

Table -III
The Effect of Ethanol extract of T. indicus on mechanical pain stimulus in albino mice

Drug	Dose (mg/ Kg b.w)	n	Reaction time in seconds (Mean ± SE)		
			30 minutes	60 minutes	90 minutes
Control (Distill water)	-	6	3.3 ±0.796	3.5 ±0.605	4.8 ±1.12
Standard (acetyl salicylic acid)	300	6	9.8 ±0.672***	8.6 ±0.886***	9.8 ±2.01
Ethanolic extract	100	6	5.2 ±0.587	5.5 ±0.509*	5.7 ±0.281
Ethanolic extract	200	6	5.3 ±0.640	5.1 ±0.237*	4.2 ±1.327
Ethanolic extract	500	6	6.1 ±0.732*	5.2 ±1.302*	4.3 ±0.847

***p<0.001 **p<001 *<0.05

Table -IV
The Effect of Ethanolic Extract of T. indicus on thermal pain stimulus in albino rats

Drug	Dose (mg/Kg b.w)	n	Reaction time in seconds (Mean ± SE)					
			Pre Drug	After30 minutes	After 60 minutes	After 120 minutes	After 180 minutes	After 240 minutes
Control (Distill water)	-	6	4.5 ± 0.730	4.83 ± 0.703	4.5± 0.671	5.0 ± 0.730	4.7 ± 0.558	5.0 ± 0.875
Standard (acetyl salicylic acid)	-	6	4.67 ± 0.558	32.66 ± 1.76***	29.34 ± 0.804***	8.67 ± 0.767***	7.83± 0.601***	6.67 ± 0.361*
Ethanolic extract	100	6	4.5 ± 0.719	7.33 ± 0.494	9.66 ± 0.496***	10.83 ± 0.6.1 ***	7.67 ± 0.334***	6.67 ± 0.422

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Ethanolic extract	200	6	5.17 ± 0.476	10.33 ± 0.615	13.67 ± 0.422***	12.16 ± 0.402***	5.83 ± 0.5	5.17 ± 0.478
Ethanolic extract	500	6	5.33 ± 0.615	11.5 ± 0.423***	12.33 ± 0.882***	12.83 ± 0.601***	7.00 ± 0.517**	6.17 ± 0.430

***p<0.001 **p<001 *<0.05

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