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### PRECLINICAL EVALUATION OF ANTI INFLAMMATORY EFFECT OF *TRAGIA PLUKENETII* R. SMITH LEAF EXTRACTS AGAINST CARRAGEENAN INDUCED PAW EDEMA IN WISTAR ALBINO RATS

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#### ABSTRACT

The aim of this study was to assess the anti-inflammatory effect of *Tragia plukenetii* R. Smith leaf extracts against carrageenan induced paw edema in wistar albino rats. The preclinical evaluation of standardized benzene, chloroform, and methanolic extracts of the leaves of *Tragia plukenetii* R. Smith was carried out for anti-inflammatory effect against carrageenan induced paw edema in wistar albino rats. The methanolic leaf extract of *Tragia plukenetii* R. Smith has shown significant anti-inflammatory effect when compared with all the other groups using carrageenan induced paw edema method. The results conclusively demonstrate the efficacy of *Tragia plukenetii* R. Smith methanolic leaf extracts for anti inflammatory activity.

#### KEYWORDS

*Tragia plukenetii*, Anti - inflammatory, Carrageenan induced paw edema and Digital plethysmometer.

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#### INTRODUCTION

The whole plant of *Tragia plukenetii* R. Smith (Family: Euphorbiaceae) is a erect, sub erect or prostrate herb, sometimes annual, up to 90 cm long, rarely more and liansscent; indumentums sparse, mostly of painful stinging hairs, distributed throughout India from Punjab and lower Himalayas eastwards to Assam and Meghalaya, ascending up to an altitude of 750 meters and southwards to Kerala<sup>1,2</sup>. Inflammation is a tissue reaction to infection, irritation or foreign substances. It is a part of the host defense mechanism. Inflammation is the complex biological response of vascular tissues to

harmful stimuli including pathogens, irritants, or damaged cells. It is a protective attempt by the organisms to remove the injurious stimuli as well as initiate the healing process for the tissue.

The process of inflammation is necessary in healing of wounds. Inflammation however if runs unchecked, lead to onset of disease like vasomotor rhinorrhoea, rheumatoid arthritis and atherosclerosis. There are several tissue factors or mechanisms that known to be involved in the inflammatory reactions such as release of histamine, that is synthesized from the amino acid histidine by the action of histidine decarboxylase, bradykinin and prostaglandins.

The inflammatory reaction is readily produced in rats in the form of paw edema (underneath the plantar region) with the help of irritants. Irritants generally known as phlogistic agents, some of them are carrageenan (sulphated polysaccharide 1% w/v), formalin (0.1ml of 1%), Bradykinin (0.1ml of 0.005%), histamine, Serotonin, mustard (0.1ml of 2.5% mustard powder suspension) or egg white (0.05ml of undiluted fresh egg white) Brewer's yeast (0.1ml of 2.5%), formaldehyde, dextran (0.1ml of 1-3% of dextran solution), kaolin (0.1ml of 5% suspension).

#### **CARRAGEENAN INDUCED PAW OEDEMA**

Carrageenan which is a phlogistic agent known to produce inflammation by releasing histamine, serotonin and bradykinin and prostaglandin. Carrageenan-induced edema is used to study the effect of drugs on acute phase of inflammation. Carrageenan is a polysaccharide obtained from seaweed (Rhodophyceae). Carrageenan induced paw edema model is known to be sensitive to cyclooxygenase inhibitors and has been used to evaluate the effect of non-steroidal anti-inflammatory agents which primarily inhibit the enzyme cyclooxygenase involved in prostaglandin synthesis<sup>3-7</sup>.

#### **EXPERIMENTAL ANIMALS**

All experimental protocols and procedures were approved by the Institutional Animal Ethics Committee of Chalapathi Institute of Pharmaceutical

Sciences. Male wistar albino rats, weighing 150-200 g, overnight fasted were used throughout the study. The animals were housed in standard laboratory conditions (12-h light/dark cycle,  $21 \pm 1^\circ\text{C}$ , and relative humidity of  $55 \pm 5\%$ ) with free access to food and water prior to the experiments. After 7 days of acclimatization to laboratory conditions, the animals were randomly assigned to experimental groups, each consisting of 5 rats. Each animal was used only once in the experimental procedures. All experiments were carried out between 9 a.m. and 3 p.m.

### **MATERIALS AND METHODS**

#### **Treatment Groups**

##### **Group-1**

Control group (0.9% normal saline 1ml/ kg, s.c.)

##### **Group-2**

Standard group (Indomethacin 20 mg/kg, s.c.)

##### **Group-3**

Benzene leaf extracts (TPBE 100mg/kg, s.c.)

##### **Group-4**

Chloroform leaf extracts (TPCE 100mg/kg, s.c.)

##### **Group-5**

Methanolic leaf extracts (TPME 100mg/kg, s.c.)

#### **Procedure**

Rats are fasted overnight with free access to water and divided into 5 groups. Rats in each group are weighed, marked with picric acid. Each group is kept in different rat cages. Both hind paw are marked with ink at the level of the lateral malleolus just beyond tibio-tarsal junction, so that the paw is dipped in the mercury column up to the fixed mark each time to ensure constant paw volume. Initial paw volume is noted by Digital Plethysmometer. Rats are divided into groups and treated into the plantar region as mentioned in treatment groups. 30 minutes later the rats are challenged by a subcutaneous injection of 0.1ml of 1% (w/v) solution of carrageenan into the plantar side of left hind paw. The right paw will serve as reference non - inflamed paw for comparison. The paw volume is measured by using Digital Plethysmometer immediately after injection, again 30, 60, 90, 120 minutes after carrageenan challenge. The difference in paw edema

volume in the right and left paw of each animal of the entire treatment group is calculated. The difference of average values between the treated animals and control groups is calculated for each time interval and statistically evaluated. The difference at the various time intervals gives some hints for the duration of the anti-inflammatory effects.

**Statistical Analysis**

All the values are expressed as mean  $\pm$ SD. Statistical significance was determined using two ways

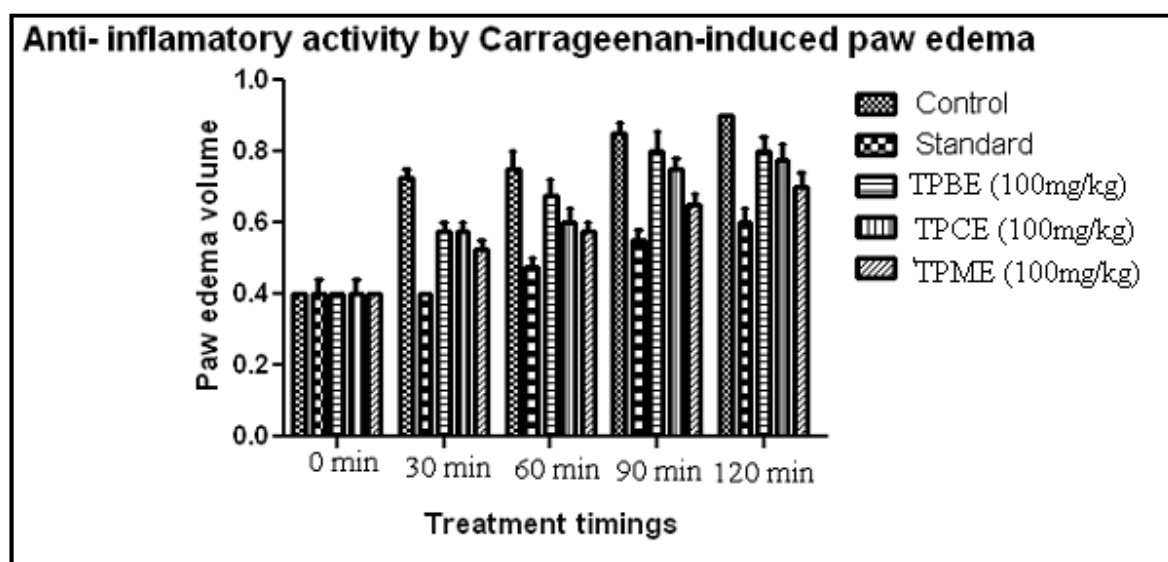
ANOVA, followed by Dunnett’s test.  $P < 0.05$  was considered to be significant.

**RESULTS AND DISCUSSION**

The methanolic leaf extracts has shown significant anti-inflammatory effect when compared with benzene and chloroform leaf extracts and control treatment groups using carrageenan induced rat paw edema method (Table No.1 and Figure No.1).

**Table No.1: Carrageenan induced rat paw edema method**

| S. No | Treatment                       | Paw edema volume (Mean $\pm$ SEM values) |                 |                 |                 |                 |
|-------|---------------------------------|--|-----------------|-----------------|-----------------|-----------------|
|       |                                 | 0 min                                    | 30 min          | 60 min          | 90 min          | 120 min         |
| 1     | Control                         | 0.4 $\pm$ 0.0                            | 0.72 $\pm$ 0.25 | 0.75 $\pm$ 0.05 | 0.85 $\pm$ 0.02 | 0.9 $\pm$ 0.0   |
| 2     | Standard (Indomethacin 20mg/kg) | 0.4 $\pm$ 0.4                            | 0.4 $\pm$ 0.4   | 0.47 $\pm$ 0.02 | 0.55 $\pm$ 0.02 | 0.6 $\pm$ 0.04  |
| 3     | TPBE (100mg/kg)                 | 0.4 $\pm$ 0.0                            | 0.5 $\pm$ 0.02  | 0.67 $\pm$ 0.05 | 0.8 $\pm$ 0.063 | 0.85 $\pm$ 0.04 |
| 4     | TPCE (100mg/kg)                 | 0.4 $\pm$ 0.04                           | 0.52 $\pm$ 0.02 | 0.57 $\pm$ 0.04 | 0.65 $\pm$ 0.02 | 0.8 $\pm$ 0.04  |
| 5     | TPME (100mg/kg)                 | 0.4 $\pm$ 0.0                            | 0.52 $\pm$ 0.02 | 0.52 $\pm$ 0.02 | 0.65 $\pm$ 0.02 | 0.7 $\pm$ 0.04  |



**Figure No.1: Differences in rat paw volume between treatment groups**

## CONCLUSION

Effectively treated animals (TPME) have shown much less edema when compared with other extract treated groups and control groups. The presence of flavonoids may be the reason for anti-inflammatory effect shown by the extracts.

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