STUDY OF ANTI-INFLAMMATORY PROPERTY OF AQUEOUS EXTRACT OF SEMECARPUS ANACARDIUM FRUIT IN RATS

Savita Patil, Kashinath Gumma, Md Mateenuddin

Department of Pharmacology, Bidar Institute of Medical sciences (Govt. of Karnataka), Bidar, KA, India

E-mail of Corresponding Author: patilsavita.884@rediffmail.com

ABSTRACT

Objectives: To evaluate aqueous extract of Semecarpus Anacardium fruit for acute and sub-acute anti-inflammatory property in albino rats and compared with the reference drug indomethacin.

Methods: For acute anti-inflammatory activity, the carrageenan induced rat paw oedema inhibition method was used and for sub-acute anti-inflammatory activity, cotton pellet granuloma method was used.

Results: It was found that percentage reduction in the paw oedema was 58.13% with Indomethacin, 48.83% and 53.48% with 100 mg/kg and 200 mg/kg Semecarpus Anacardium extract. Reduction in the granuloma formation was 60.2% with Indomethacin, 54.7% and 56% with 100 mg/kg and 200 mg/kg Semecarpus Anacardium extract respectively.

Conclusion: Aqueous extract of Semecarpus Anacardium fruit showed significant anti-inflammatory effect.

Keywords: Semecarpus Anacardium fruit extract; anti-inflammatory; Rat paw oedema method; Cotton pellet granuloma method

INTRODUCTION

Inflammatory diseases are one of the major causes of morbidity. Inflammation is the dynamic process by which living tissue reacts to injury [1]. Though standard drugs like aspirin, indomethacin are available but these drugs are not entirely free from side effects and have their own limitations [2,3]. It is believed that current analgesia inducing drugs such as opiates and NSAIDS are not useful in all cases, because of their side effects like gastrointestinal irritation, liver dysfunction and many others [4]. Thus there is still a need to develop newer and safer anti-inflammatory drugs. Semecarpus Anacardium (Family: Anacardiaceae) also called the “marking nut” has found many applications in Indian medicine in the treatment of gout, rheumatic pain and cancer [5]. A variety of nut extract preparations of semecarpus are effective against many diseases like arthritis, tumours, infections etc and non toxic even at high dose of 2000mg/kg [6]. However systematic study of this plant has not been carried out for these properties. The present study evaluates the anti-inflammatory activity of Semecarpus Anacardium fruit in albino rats.

MATERIALS AND METHODS

Preparation of extract: The fruits of Semecarpus anacardium were shade dried and powdered. Subsequently aqueous extract was obtained using maceration process. The extract obtained was used for oral administration in albino rats.

Selection of animals, caring and handling: Healthy Wistar rats (150–200 g), aged twelve weeks of either sex, bred locally in the animal house of Bidar Institute of Medical Sciences,
Bidar were selected for the study. They were housed under the temperature of 23±2°C, relative humidity of 30–70% and 12 h light–12 h dark cycle. All animals were fed with standard diet and had free access to water. The study was done after obtaining approval of Institutional Animal Ethics.

**Study design**

**Carrageenan induced rat paw oedema inhibition:** Albino rats were divided into 4 groups, each containing 6 rats.

- **Group I:** Control treated with normal saline per orally (0.1 ml).
- **Group II:** Indomethacin (20 mg /kg) p.o.
- **Group III:** Test group A: Aqueous extract of Semecarpus anacardium fruit (100 mg/kg) p.o.
- **Group IV:** Test group B: Aqueous extract of Semecarpus anacardium fruit (200 mg/kg) p.o.

Acute inflammation was produced by injecting 0.1ml of 1% carrageenan suspension in normal saline into the subplantar region of right hind paw after 30 minutes of drug administration. A mark was made on the leg at the malleous to facilitate uniform dipping at subsequent readings. The volume of paw oedema volume was measured with the help of plethysmograph by mercury displacement method immediately before and three hours after the drug administration. The percentage inhibition of oedema in various treated groups was then calculated by using statistical analysis.

**COTTON PELLET GRANULOMA METHOD**

Rats were divided into 4 groups, each group containing 6 rats. Under ether anaesthesia, the the axillary and groin region hairs were cut and sterile cotton pellets of 10mg each were implanted in the subcutaneous tissue on either sides of axilla and sterile grass pith (25 x 2mm) in the groin region. Wounds were then sutured and animals were caged individually after recovery from anaesthesia. The rats then received treatments as described earlier. The test drug administration was started on the day of implantation and given daily for 7 days. During this period any change in the behaviour like food intake, motor activity and diarrhoea, were noted.

On the 8th day, the rats were sacrificed and cotton pellets and grass piths removed. The pellets free from the tissue were dried overnight. Net granuloma formation was determined by subtracting the initially weight noted (i.e.10 mg).

**Statistical analysis:** One factor ANOVA followed by Newman – Keul’s studentized range test was used for comparing with control. Fisher’s exact test was used to compare ulcer incidence.

**RESULTS**

**Effect on carrageenan induced paw edema inhibition test (Table -1)**

In carrageenan induced rat paw edema test, the doses of 100mg/kg and 200 mg/kg aqueous extract of Semecarpus Anacardium fruit showed statistically significant (P<0.05) inhibitory effect on “mean increase in paw volume”. Semecarpus Anacardium extract showed acute anti inflammatory activity higher than control group, but it did not show so strong effect as Indomethacin, which produced significant inhibition (58.13%) (P<0.05). It was found that reduction in the inflammation was 48.83 % (P<0.05) with 100mg/kg Semecarpus Anacardium fruit extract and 53.48 % (P<0.05) with 200 mg/kg Semecarpus Anacardium fruit extract.

**Effect on Cotton pellet granuloma inhibition method: (Table -2)**

The dry weight of cotton pellet granuloma in control, two different doses of Semecarpus Anacardium fruit extract and Indomethacin treated groups is shown in the Table 2. It can be noted that the two doses of Semecarpus Anacardium fruit extract 100 mg/kg and 200 mg/kg, and Indomethacin showed significant (P<0.05) activity in inhibiting dry weight of granuloma. The extract administered at 200 mg/kg, p.o. had a greater anti-
granulation (56%) effect but less than indomethacin (60.2%). Semecarpus Anacardium fruit extract also showed lower percentage of ulcers compared to indomethacin. (Table -3)

DISCUSSION
In present study extract of Semecarpus Anacardium fruit extract at a dose of 100mg/kg & 200mg/kg showed 48.83% & 53.48% inhibition (p < 0.05) of rat-paw oedema respectively. Efficacy of anti-inflammatory agents in sub acute inflammatory states is indicated by their ability to inhibit the increase in the number of fibroblasts and synthesis of collagen and mucopolysaccharides during granuloma tissue formation [7]. Semecarpus Anacardium fruit showed significant (P <0.05) anti-inflammatory activity by reducing granulomatous tissue in cotton pellet granuloma method and thus found to be effective in sub acute inflammatory condition. Semecarpus Anacardium extract also showed lower percentage of ulcers compared to indomethacin. Thus, this could be an added advantage with respect to other anti-inflammatory drugs. On the basis of these findings, it may be inferred that aqueous extract of Semecarpus Anacardium fruit has acute and sub acute anti-inflammatory activities. At present, there are less reports on investigation to identify the active components present in aqueous extract of Semecarpus Anacardium fruit. Further investigations are anticipated to identify the active components and lead to their further clinical use.

CONCLUSION
On the basis of present study it can be concluded that aqueous extract of Semecarpus anacardium fruit seem to possess anti-inflammatory property.

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REFERENCES
Table 1: Showing oedema volumes (ml) in carrageenan induced rat paw oedema inhibition test

<table>
<thead>
<tr>
<th>Groups</th>
<th>Dose/route</th>
<th>Paw oedema ml (%)</th>
<th>Inhibition %</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Mean ± S.E.M</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group I</td>
<td>Control</td>
<td>0.1ml p.o.</td>
<td>0.43 ± 0.30</td>
<td>---</td>
</tr>
<tr>
<td>Group II</td>
<td>Indomethacin</td>
<td>20 mg/kg p.o.</td>
<td>0.18 ± 0.33</td>
<td>58.13</td>
</tr>
<tr>
<td>Group III</td>
<td>Test group A</td>
<td>100mg/kg p.o.</td>
<td>0.22 ± 0.30</td>
<td>48.83</td>
</tr>
<tr>
<td>Group IV</td>
<td>Test group B</td>
<td>200 mg/kg p.o.</td>
<td>0.20 ± 0.36</td>
<td>53.48</td>
</tr>
</tbody>
</table>

p.o: per orally,  S.E.M: Standard error of mean, Test group A and B = Aqueous extract of semecarpus anacardium fruit
*P < 0.05: Significant with respect to control group

Table 2: Weight of the dry cotton pellet in the cotton pellet granuloma method

<table>
<thead>
<tr>
<th>Groups</th>
<th>Dose/route</th>
<th>Weight of dry cotton pellet granuloma (mg)</th>
<th>Inhibition %</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Mean ± S.E.M</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group I</td>
<td>Control</td>
<td>0.1ml p.o.</td>
<td>18.27 ± 0.32</td>
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<tr>
<td>Group II</td>
<td>Indomethacin</td>
<td>20 mg/kg p.o.</td>
<td>7.27 ± 0.18</td>
<td>60.20</td>
</tr>
<tr>
<td>Group III</td>
<td>Test group A</td>
<td>100 mg/kg p.o.</td>
<td>8.27 ± 0.34</td>
<td>54.70</td>
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<tr>
<td>Group IV</td>
<td>Test group B</td>
<td>200 mg/kg p.o.</td>
<td>8.03 ± 0.27</td>
<td>56</td>
</tr>
</tbody>
</table>

p.o: per orally,  S.E.M: Standard error of mean, Test group A and B = Aqueous extract of semecarpus anacardium fruit
*P < 0.05: Significant with respect to control group

Table 3: Ulcer incidence of indomethacin and test drug

<table>
<thead>
<tr>
<th>Groups</th>
<th>Ulcer incidence</th>
<th>P* Value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Rats with ulcer</td>
<td>With Control</td>
</tr>
<tr>
<td>Group I</td>
<td>Control</td>
<td>2 (6)</td>
</tr>
<tr>
<td>Group II</td>
<td>Indomethacin</td>
<td>6 (6)</td>
</tr>
<tr>
<td>Group III</td>
<td>Test group A</td>
<td>3 (6)</td>
</tr>
<tr>
<td>Group IV</td>
<td>Test group B</td>
<td>4 (6)</td>
</tr>
</tbody>
</table>

*P < 0.05: Significant,  *P > 0.05: Not Significant, Test group A and B = Aqueous extract of semecarpus anacardium fruit