COMPARATIVE STUDY ON ANTI-INFLAMMATORY ACTIVITY OF CYPERUS RODUNTUS (L.) USING DIFFERENT SOLVENT SYSTEM IN CARRAGEENAN INDUCED PAW EDEMA IN ALBINO WISTAR RATS

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ABSTRACT

Cyperus rotundus is a perennial plant that may reach a height of up to 40 cm. It vernacularly called “Nagarmotha” is a medicinal plant belonging to the family of the Cyperaceae and appearing among Indian, Chinese, Japanese natural drugs. Used as home remedy against spasms, stomach disorders and irritation of bowl. In Indian System of medicine, the rhizome of the plant has been recommended for use in several Clinical conditions like fever and arthritis. Albino Wistar rats weighing about 150-180 gm were divided into eight groups six no in a group and 1 % v/v of carrageenan injected to all the animal in the groups and their Paw volume was measured initially at intervals of 15, 30, 60, 120, 180 mins. After carrageenan was injected by volume displacement method using Plethysmometer by immersing the paw into the mercury cell the edema volume measured, followed with administration of test drug to the Group 1 (control) animals 1% v/v Tween 80 (0.2 ml/kg, p.o.), Group II (standard) received Indomethacin (10 mg/kg, p.o.), Group III (Test) received 200, 400 mg/kg dose of of Aqueous extract of Cyperus roduntus, Group IV received 200, 400 mg/kg ether extract of Cyperus roduntus, Group V received 200, 400 mg/kg Ethanol extract of Cyperus roduntus respectively. In this study the given test drug extract of Cyperus roduntus had favourably produced maximum % inhibition of paw edema at 400 mg/kg dose of each solvent extraction in the grouped animal models as equal to the standard drug indomethacin. The present study demonstrated anti-inflammatory effect of Ethanolic extract of Cyperus roduntus was equal to standard anti-inflammatory drug (Indomethacin).

Keywords: Cyperus roduntus, Indomethacin, Carrageenan, Anti-inflammatory.

INTRODUCTION

Cyperus rotundus L. (Family: Cyperaceae, Koraikizangu in Tamil, Thunga in Telugu, Dutgrass in English) is a perennial sedge distributed throughout India. The tubers are externally blackish in colour and reddish white inside, with a characteristic odour. The stems grow to about 25 cm tall and the leaves are linear, dark green and grooved on the upper surface. Inflorescences are small, with 2-4 bracts, consisting of tiny flowers with a red-brown husk. The nut is three-angled, oblong-ovate, yellow in colour and black when ripe. C. rotundus is indigenous to India, but are now found in tropical, subtropical and temperate regions (Pooley E, 1998; Gordon-Gray KD, 1995).

Roots and tubers of this plant are used in different diseases like chronic diarrhea, inflammation, skin rashes and excess bleeding. It has also antiestrogenic, antimicrobial, anathematic, antihistaminic, antiemetic, antipyretic, anti diabetic and antioxidant activities (Pal DK and Dutta S, 2006; Weenam H et al., 1990).
Cyperus rotundus is a multipurpose plant, widely used in traditional medicine around the world to treat stomach ailments, wounds, boils and blisters (Oliver-Bever B, 1986; Puratuchikody A et al., 2006; Joshi AR and Joshi K, 2000; El-Kamali HH and El-Khalifa KP, 1999; Durate MCT et al., 2005). A number of pharmacological and biological activities including anti-Candida, anti-inflammatory, anti-diabetic, anti-diarrhoeal, cytoprotective, antimutagenic, antimicrobial, antibacterial, antioxidant, cytotoxic and apoptotic, anti-pyretic and analgesic activities have been reported for this plant (Durate MCT et al., 2005; Sundaram MS et al., 2008; Raut NA and Gaikwad NJ, 2006; Uddin SJ et al., 2006 Kilani S et al., 2005; Zhu M et al., 1997; Kilani S et al., 2007& 2008; Dhillon RS et al., 1993; Pal DK and Dutta S, 2006; Oladipupo A Lawal and Adeoba Oyedeji, 2009). Previous phytochemical studies on C. rotundus revealed the presence of alkaloids, flavonoids, tannins, starch, glycosides and furochromones, and many novel sesquiterpenoids.

MATERIALS AND METHODS

Plant collection

The tubers of the Cyperus rotundus were collected from North Rajupalem, Nellore District, Andhra Pradesh, India. The collected tubers were identified and authenticated by Prof. Jayaraman, Director, National institute of Herbal science, Tambaram, Chennai, Tamil Nadu, India. A Voucher specimen (PARC/ 2011 / 1040) was obtained on 21.12.2011 and deposited in Herbarium of our department. The tubers were shade dried at room temperature for 15 days and make in to coarse powder.

Preparation of Extracts by Maceration Process

The plant tubers collected from the local place were made into fine powder by grinding procedure, after that it was divided into three groups carrying 150 g each, followed with addition of 3 / 4 volume of Ether 90 %, Ethanol (99 %), Distilled water Individually with different solvent system. They were immersed completely by Maceration Process and allowed to wait for five days. Finally it was filtered by muslin cloth and the solvents were completed evaporated by hot plate method finally collected the dried residue for further activity.

Animals

Adult Albino Wistar rats weighing about 150-180 g of either sex were procured from the animal house. The animals were maintained in a well-ventilated animal house approved by Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), with 12:12 hour light/dark cycle in polypropylene cages with 27±2°C temperature. The animals were given standard diet.

Drugs and chemicals used

Aqueous extract of Cyperus rotundus (L.), Ether extracts of Cyperus rotundus (L.) (EtCR) Ethanolic extract of Cyperus rotundus (L.) (ECR), Carrageenan, Indomethacin (10 mg/kg), 1% v/v Tween 80.

Procedure for induction of inflammation

The anti-inflammatory activity of the extracts was determined using Carrageenan induced rat paw edema assay (Winter et al., 1962). After 30 mins of the treatment, 0.1ml of 1% carrageenan in saline was injected into the sub plantar region of the left hind paw of each rat to induce edema. The paw volume was measured initially and at intervals of 15, 30, 60, 120, 180 mins. After carrageenan injection by volume displacement method using Plethysmometer by immersing the paw in mercury cell. The percentage inhibition of paw volume in drug treated group was compared with control group. Indomethacin (10 mg/kg) was used as standard drug. The percentage inhibition of paw edema was calculated by using the following formula;

\[
\text{Percentage of edema inhibition} = \left(\frac{V_c - V_t}{V_c}\right) \times 100
\]

Vc- Volume of paw edema in control group
Vt- volume of paw edema in treated group

Experimental Design

The animals were weighed, Numbered and separated as 8 groups as per dose as (n = 6) six animals (albino wistar rat) per group and different dose of control drug, standard drug, test drugs were administered as the same to all group of animals.

- Group I : 1% v/v Tween 80 (0.2 ml/kg, p.o.) [Negative Control Group]
- Group II : Indomethacin (10 mg/kg, p.o) [Standard group]
- Group III : 200 mg/kg of ACR suspended in 1% v/v Tween 80, p.o.
- Group IV : 400 mg/kg of ACR suspended in 1% v/v Tween 80, p.o.
- Group V : 200 mg/kg of EtCR suspended in 1% v/v Tween 80, p.o.
- Group VI : 400 mg/kg of EtCR suspended in 1% v/v Tween 80, p.o.
- Group VII : 200 mg/kg of ECR suspended in 1% v/v Tween 80, p.o.
- Group VIII : 400 mg/kg of ECR suspended in 1% v/v Tween 80, p.o.

Statistical analysis

The statistical analysis was carried out using Graph pad prism 4.0 software. All values were expressed as Mean ± S.E.M. Data analysis was done by one-way ANOVA followed by Dunnett’s multiple comparison tests. Difference level at P≤0.05 was considered as statistically significant condition. Result were considered to be significant if p < 0.01 and highly significant if p < 0.001.
RESULTS

Table 1. Effect of *Cyperus roduntus* L. on Carrageenan Induced paw edema

<table>
<thead>
<tr>
<th>Groups</th>
<th>Paw edema volume (ml)</th>
<th>% Inhibition of paw volume</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0min</td>
<td>15min</td>
</tr>
<tr>
<td>Group I (Negative Control)</td>
<td>0.80±0.01</td>
<td>1.06±0.01</td>
</tr>
<tr>
<td>Group II (standard)</td>
<td>0.82±0.02</td>
<td>1.12±0.02*</td>
</tr>
<tr>
<td>Group III (ACR-200)</td>
<td>0.80±0.01</td>
<td>1.21±0.02*</td>
</tr>
<tr>
<td>Group IV (ACR-400)</td>
<td>0.81 ±0.01</td>
<td>1.14±0.02*</td>
</tr>
<tr>
<td>Group V (EtCR-200)</td>
<td>0.77 ±0.01</td>
<td>1.17±0.01*</td>
</tr>
<tr>
<td>Group VI (EtCR-400)</td>
<td>0.78 ±0.02</td>
<td>1.13±0.02*</td>
</tr>
<tr>
<td>Group VII (ECR-200)</td>
<td>0.76 ±0.01</td>
<td>1.17±0.02*</td>
</tr>
<tr>
<td>Group VIII (ECR-400)</td>
<td>0.84 ±0.01</td>
<td>1.03±0.02*</td>
</tr>
</tbody>
</table>

*p < 0.01, **p < 0.001 When compared with control (One way ANOVA test)

Fig: 1 (a) Effect of *Cyperus roduntus* L. on carrageenan induced paw edema
DISCUSSION

Cyperus roduntus is the perennial plant which grows at Summer and Winter season, collected and used for the present study since its containing high amount of active chemical constituents in it which was favorable for anti-inflammatory activity [20]. Thereafter the plant tubers was powdered by grinding method in the laboratory followed by it was separated into a three heaps which was separated as 150 gm each. Under the maceration process the powder was soaked into different solvents individually in a separate beakers containing Aqueous solvent (Distilled water), Ether solvent, Ethanol solvent was kept aside in the laboratory with daily care by the students consecutively five days, thereafter it was filtered by muslin cloth and the retaining solvent was completely evaporated on the hot plate evaporation method.

Edema is induced by using 1% v / v of indomethacin injection at the articular joint and followed by administration of Test drug to the different groups of animals. Cyperus roduntus plant extract in the paw oedema test had shown desirable effect when the test samples ACR (group III) 200 mg / kg , 400 mg / kg (group – IV ), and EtCR 200mg / kg , 400 mg / kg, and ECR 200 mg / kg , 400 mg / kg animals were compared with Standard group Indomethacin (Group II) , each individual group used six (n=6 ) no. of animals to compare the anti-inflammatory activity. The time were scheduled at intervals of 0 min, 15 , 30 , 60 , 120 , 180 min to measure the edema volume of the each wistar rats after the carrageenan injected into animals through articular joint.

When compared to the indomethacin (standard group) paw edema % of inhibition 67.57% against Test ACR 200 mg/kg = 60 % and 400 mg/ kg = 61.62% the aqueous system had partial inhibition of paw edema whereas EtCR 200=61.62 %, 400 mg/ kg = 63.78% which was shown alternatively good paw oedema inhibitory action after 180 min. it has standard deviation of 0.67 ±0.01** when compared with indomethacin standard deviation values 0.60 ±0.02** with the help of ANOVA method. The ECR group animals 200 mg/ kg = 62.70% , 400 mg/kg = 65.40% were almost produced same action as standard group animals and the standard deviation was 0.64 ±0.02** *p < 0.01, **p < 0.001,When its compare to the different solvent system the maximum % inhibitory action potential revealed by ECR :400 mg / kg dose against control and standard groups. The ECR group 400 mg / kg dose produced best % inhibitory action of paw edema which was induced by carrageenan related to all other solvent system preferably dose 400 mg / kg, since it has high rate of miscible of different chemical constituent in it compare to EtCR and ACR solvent system.

CONCLUSION

Cyperus roduntus is a potent anti-inflammatory drug to cure the inflammation in carrageenan induced inflammation on the albino wistar rats, when the different
doses of the plant solvent extract was compared against the standard drug indomethacin 10 mg / kg. The Ethanolic extract dose 400 mg / kg was shown good anti-inflammatory effect against other solvent system, since high amount of favorable active constituent present in it.

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