EVALUATION OF ANTI-INFLAMMATORY ACTIVITIES OF EXTRACT OF LEAVES OF ECLIPTA ALBA AND WEDELIA CALENDULACEA

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ABSTRACT
The present study is the evaluation of anti-inflammatory activity of the various extracts of the leaves of Eclipta alba L. and Wedelia calendulacea. The anti-inflammatory activity of aqueous, hydro alcoholic and alcoholic extract of Eclipta alba (1kg/mg p.o.) and Wedelia calendulacea (1kg/mg p.o.) were carried out by using carrageenan-induced paw oedema model. Either sex of S.D. rats was used. Indomethacin was used as a standard drug (positive control). The alcoholic extract significantly inhibited the carrageenan-induced paw oedema at 3rd hr as compare to aqueous and hydro alcoholic extract. The present study indicates that the oral administration of alcoholic extract of both the plant showed anti-inflammatory activity.

Keywords: Eclipta alba, Wedelia calendulacea and anti-inflammatory.

INTRODUCTION

Inflammation is a very complex, dynamic, multifactored phenomenon involving many systems. It is closely interwoven with the process of repair. Although inflammation and repair are basically defense mechanisms they are potentially harmful. No known anti-inflammatory drugs are equally effective in suppressing all the facets of inflammatory reactions. The available NSAIDs agents do not modify the progression of the rheumatic disorder they offer symptomatic relief making life easier to the afflicted person. Hence, they have important therapeutic role and are used on large scale. Unfortunately their usage is produced many of the adverse effect like ulceration is stomach and duodenum, nephrotoxicity especially in patients who already suffer from the renal insufficiency. Hematological disturbance like inhibit platelets aggregation. In the early development of modern medicine biologically active compounds from higher plants have played a vital role in providing a medicine to combat pain and disease [¹].
The plant *Eclipta alba* L. Hassk. (syn. *E. prostrate* L., Asteraceae) and *Wedelia calendulacea* popularly known as “Bhangra” in English, and "Bringaraja" in Sanskrit, with white and yellow flower respectively is a common remedy for various ailments. It has been cultivated as a common weed throughout India, particularly in cool and moist places. The plant contains coumestans i.e. wedelolactone (I) and demethylwedelolactone (II), polypeptides, polyacetylenes, thiophene-derivatives, steroids, triterpenes, flavonoids and nicotine [2, 3, 4, 5]. Pharmacologically *Eclipta alba* was used as a hepatoprotective [5], antiviral [6], anti-analgesic [7], anti-epileptic [8], Anti-Oxidants [9], Inhibiting the IKK Complex [10], Immunomodulatory activities [11]. Pharmacologically *Wedelia calendulacea* was used as an Immunostimulant [12], antibacterial [13], wound healing [14] and Anticancer [15].

In the present study, sincere efforts has been attempted to establish the scientific validity of the anti-inflammatory activity of the leaves of both the plants.

**MATERIALS AND METHODS**

**Plant Material**

The fresh leaves of plant *Eclipta alba* and *Wedelia calendulacea* were collected from the area of Sanand village, Nr. Ahmedabad, GUJARAT, INDIA, in November-December. The herbarium of this plant was identified and authenticated [Herbarium No. 1342/144] by Dr. H. B. Singh, Scientist F & Head, Raw Materials Herbarium & Museum, NISCAIR, New Delhi, INDIA. The leaves were cut into small pieces and shade dried at room temperature, dried leaves was subjected to size reduction to a coarse powder by using a dry grinder.

**Chemicals**

Kappa Carageenan was purchased from Purvi Enterprise, Ahmedabad and Indomethacine from the market preparation. All the solvents used for the extraction were of AR grade.

**Preparation of Plant Extract**

The coarse powder of the leaves (250 g) was extracted in soxhlet extraction with the sufficient quantity of Water, Methanol : Water (50:50) and Methanol for 24 hours. Solvent was removed under reduced pressure at 40°C. Extracts were stored in an airtight
container in refrigerator, which was further suspended in 1% w/v tween-80 for the oral administration.

Selection of Animal

Sprague Dawley rats of either sex of weighing 250-300g were procured from central animal facility of L. J. Institute of Pharmacy, Ahmedabad. They were fed with the standard food pellets (Pranav agro, Baroda) and water ad libitum. They were housed in polypropylene cages maintained under standard conditions.

Ethics

The experimental protocol was subjected to the scrutiny of the Institutional Animal Ethics Committee, and was cleared by same before beginning the experiment (No. LJIP/IAEC/01/2011–2012).

Carrageenan Induced Paw Edema

Acute inflammation was produced in all animals by sub plantar injection of 0.1 ml freshly prepared suspension of 1% Carrageenan in normal saline on the left hind paw of rats. Paw thickness was measured using a plethysmograph before and after Carrageenan challenge in each group. Animals were premeditated with extract of plant drugs and the std. drug, Indomethacine, orally 1 h before Carageenan injection. In all the above models, the degree of edema formation was determined as increase in paw thickness. Increase in paw thickness and percent inhibition was calculated as under.

\[
\text{Increase in Paw Volume} = V_t - V_0
\]

Where, \( V_t \) = Paw Volume at t Time, \( V_0 \) = Paw Volume at 0 Time

\[
\% \text{ Inhibition in paw volume} = \left( \frac{V_c - V_t}{V_c} \right) \times 100
\]

Where, \( V_c \) = Increase in Paw Volume in Control, \( V_0 \) = Increase in Paw Volume in Treatment

Group of Animal:

Group – I : Disease Control animals treated with 1% w/v tween-80, orally
Group – II : Aqueous extracts of Eclipta alba 1 gm/kg orally
Group – III : hydro alcohohic extracts of Eclipta alba 1 gm/kg orally
Group – IV : Alcoholic extracts of Eclipta alba 1 gm/kg orally
Group – V : Aqueous extracts of Wedelia calendulacea 1 gm/kg orally
Group – VI :  hydro alcoholic extracts of Wedelia calendulacea 1 gm/kg orally
Group – VII :  Alcoholic extracts of Wedelia calendulacea 1 gm/kg orally
Group – VIII :  Positive control Indomethacine 10 mg/kg orally

Each group having 6 animals

Statistical Analysis

The results were expressed as mean ± S.E.M. Statistical difference between two means determined by unpaired t-test using software Graph pad prism.

RESULTS

Carrageenan Induced Paw Edema

In acute inflammation model, the EtoAc/Me-OH extract inhibited carrageenan-induced paw oedema by 61.92 ± 2.18 % at a dose of 250mg/kg, 76.12 ± 2.65% at a dose of 500mg/kg and 74.61 ± 1.50% at a dose of 750mg/kg. The Me-OH/Me-OH extract inhibited carrageenan-induced paw oedema by 64.46 ± 3.66 % at a dose of 250mg/kg, 83.24 ± 1.71% at a dose of 500mg/kg and 83.75 ± 1.50% at a dose of 750mg/kg. The std. indomethacin inhibits carrageenan-induced paw oedema by 88.32 ± 1.45% at the dose of 10mg/kg P.O. The effects of 500mg/kg of both the extract were comparable to effect produced by std. indomethacin.

**TABLE 1: EVALUATION OF ANTI-INFLAMMATORY ACTIVITY OF VARIOUS LEAF EXTRACT OF ECLIPTA ALBA ON CARAGEENAN INDUCED PAW EDEMA MODEL**

<table>
<thead>
<tr>
<th>Group</th>
<th>Paw edema (Vol. in ml)</th>
<th>% Inhibition</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1 hr</td>
<td>2 hr</td>
</tr>
<tr>
<td>I</td>
<td>0.33 ± 0.054</td>
<td>0.40 ± 0.076</td>
</tr>
<tr>
<td>II</td>
<td>0.42 ± 0.017*</td>
<td>0.39 ±0.015</td>
</tr>
<tr>
<td>III</td>
<td>0.31 ± 0.021**</td>
<td>0.27 ± 0.017*</td>
</tr>
<tr>
<td>IV</td>
<td>0.35 ± 0.020***</td>
<td>0.26 ± 0.016**</td>
</tr>
<tr>
<td>V</td>
<td>0.42 ± 0.017</td>
<td>0.39 ± 0.015*</td>
</tr>
<tr>
<td>VI</td>
<td>0.31 ± 0.021</td>
<td>0.27 ± 0.017**</td>
</tr>
<tr>
<td>VII</td>
<td>0.35 ± 0.020</td>
<td>0.26 ± 0.016**</td>
</tr>
<tr>
<td>VIII</td>
<td>0.28 ± 0.054**</td>
<td>0.16 ± 0.033**</td>
</tr>
</tbody>
</table>

* P<0.05, ** P<0.01, *** P<0.001 as compare to control (Paw edema) and to positive control (% Inhibition) as per student’s t-test. Values are expressed in Mean ±S.E.M. n=6 animal in each group.
DISCUSSION

The anti-inflammatory activity of plant extract was evaluated using carrageenan induced paw edema models of inflammation. Carrageenan-induced paw oedema as an in vivo model of inflammation was selected to assess the anti-inflammatory activity of natural product and as an acceptable and reliable method for acute anti-inflammatory studies [17]. Oedema formation due to carrageenan in the rat is a biphasic event. The first phase (0-1 hour after Carrageenan) mainly results from concomitant release of inflammatory mediators, namely histamine, serotonin and kinin from surrounding the damaged tissue. The second phase (1-3 hours after Carrageenan) is mediated by bradykinin, leukotrienes and sustained release of prostaglandins produced by the macrophages. The second phase is sensitive to most clinically effective anti-inflammatory drugs [18].

Oral administration of the high dose of alcoholic extract of Eclipta alba and Wedelia calendulacea suppressed the edematous response after 2 h and this effect continued upto 3 h as compare to aqueous and hydro alcoholic extract of both the plants. The observed effect was similar to that of std. indomethacin. NSAIDS block the synthesis of prostaglandins by inhibiting cyclooxygenase. Naturally occurring polyphones such as flavonoids, coumarins and tannin might be expected to interfere with the process of synthesis of prostaglandins to produce anti-inflammatory effects [19].

The results of the present study indicated that alcoholic extract of Eclipta alba and Wedelia calendulacea significantly inhibited the formation of rat hind paw edema in the late phase. This effect may be due to influence on inflammatory mediators and also on pathway of prostaglandins synthesis which may be due to the presence of flavonoids, coumarins and other polyphenolic compounds in plant extract. However the chemical constituents and mechanism responsible for the pharmacological activities remain investigated.

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REFERENCES


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