ANTI-ULCEROGENIC ANTI-INFLAMMATORY ACTIVITY OF
BXUS WALLICHIANA.

N. R. Livingston Raja, G.Vinatha Pooshan, Neethu Joseph, Dayanajose.

Arulmigu Kalasalingam College of Pharmacy, Krishnankoil, Tamilnadu, India – 626 190.

Summary

The various extracts were prepared from Buxus Wallichiana. The anti-inflammatory activity of various doses of petroleum ether, chloroform and methanol extracts were evaluated in rats. The percentage of edema inhibition was found better in petroleum ether extract than other extracts. It was concluded that anti-inflammatory activity of petroleum ether extract of Buxus Wallichiana is mediated through three phase mechanism.

Key words: Buxus Wallichiana, inflammatory, edema, chloroform.

Correspondenceauthor; G. Vinatha Pooshan, E.mail: g_pooshan@yahoo.com

Introduction

In general, the traditional anti-inflammatory drugs both steroidal as well as non-steroidal are mostly associated with test of unwanted side effects including ulceration which are the most common and a serious problem[1]. Plants are the most important source for the new drug development due to its safe use. Our aim of the present study is to access the possible anti ulcerogenic—anti inflammatory activity of the various extract of the plant Buxus Wallichiana. Plants are the most important source for New Drug development due to its safe use. Our aim of the present study is to access the possible anti ulcerogenic—anti inflammatory activity of various extract of the plant of Buxus Wallichiana.

Materials and Methods

Buxus wallichiana leaves were collected from the Doddapetta region of Nilgiris dist, Tamil Nadu, India, during the month of march of 2009. The plant was identified and authenticated by Dr Steefen, Department of Botony, Kamaraj University Madurai.
Preparation of Extract

Fresh *Buxus Wallichiana* leaves were collected, shade dried and powdered mechanically. About 100g of the leaf powder extracted with 600ml of petroleum ether, chloroform and methanol by hot percolation process. The extracts were dried at 40°C under vacuum and the yield of the extracts were found to be 34, 24, 30, 37%w/w. The yield extracts were freshly prepared and used for the pharmacological studies.

Animals

Male albino rats of Wister Strain weighing between 150gm and 200gm were used. They were housed in standard cages at room temperature (25±2°C) and provided with food and water. The study was conducted after obtaining institutional ethical committee clearance bearing No 509/02/CPCSFA/2002.

Statistical Analysis

Statistical analysis was performed using unpaired two tailed students t test. The values were considered significant the level’s of p<0.05 and p<0.01.

Methodology

**Carrageenan Induced Paw edema model [2]**

**Experimental set up**

Male Albino rats of Wister strain weighing about 150-200gm were divided into seven groups of five animals each and the extract were administered orally at a dose of 300; 600mg per kg  
GROUP 1: Indomethacin 10mg per kg (std) is administered orally  
GROUP 2:PEBW 300mg per kg is administered orally  
GROUP 3:PEBW 600mg per kg is administered orally  
GROUP 4:CEBW 300mg per kg is administered orally  
GROUP 5:CEBW 600mg per kg is administered orally  
GROUP 6:MEBW 300mg per kg is administered orally  
GROUP 7:MEBW 600mg per kg is administered orally
**Procedure**

1. Wistar Albino rats were used for the study and the extracts were administered for eight days orally.  
2. To the right hind paw (sub plantar region) of the rat, 0.1ml of 1% carrageenan (suspended in normal saline) was injected subcutaneously on 8th day, after that per oral administration of the diclofenac sodium (std) and extracts (test) were given to the above groups respectively.  
3. The paw volume was measured by using plethysmometer (UGO Baisle ; Italy) at the time interval of 0.5, 1, 2, 3 and 5 hours.  
4. The difference between the initial paw volume and after drug administer paw volume were considered as change in paw volume.  
5. The % of protection was calculated by the following formula.

\[
\%\text{ protection} = \frac{\text{control} - \text{treatment}}{\text{control}} \times 100
\]

**Ulceregenic Potential**

At the end of the experiment (day 8) in carrageenan induced paw model, each group of drug (six hours after drug) were sacrificed and inflated with normal saline and soaked in 10% formalin for 30 minutes.[6]

1. After which the stomach was cut along the greater curvature and washed with saline for any adherents.  
2. The stomach was pinned in a black wooden board and the ulcer score was taken visually.

The ulcerogenic potential was assessed as described before.  
0: No visible erosion (or) blood.  
1: Blood in stomach (or) one intermediate erosion.  
2: Several pin point (or) one intermediate erosion.  
3: Several intermediate (or) one large erosion.  
4, 5, 6: progressively greater mucosal erosion results.

**Results**

**Table1: % Inhibition of edema**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Dose</th>
<th>0.5</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Indomethacin</td>
<td>10mg/kg</td>
<td>40</td>
<td>43</td>
<td>57</td>
<td>75</td>
<td>82</td>
</tr>
<tr>
<td></td>
<td>300mg/kg</td>
<td>46</td>
<td>50</td>
<td>71</td>
<td>72</td>
<td>56</td>
</tr>
<tr>
<td>PEBW</td>
<td>300mg/kg</td>
<td>40</td>
<td>46</td>
<td>48</td>
<td>54</td>
<td>52</td>
</tr>
<tr>
<td></td>
<td>600mg/kg</td>
<td>46</td>
<td>50</td>
<td>71</td>
<td>72</td>
<td>56</td>
</tr>
<tr>
<td>CEBW</td>
<td>300mg/kg</td>
<td>15</td>
<td>25</td>
<td>33</td>
<td>27</td>
<td>14</td>
</tr>
<tr>
<td></td>
<td>600mg/kg</td>
<td>34</td>
<td>45</td>
<td>55</td>
<td>50</td>
<td>44</td>
</tr>
<tr>
<td>MEBW</td>
<td>300mg/kg</td>
<td>32</td>
<td>45</td>
<td>49</td>
<td>25</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>600mg/kg</td>
<td>40</td>
<td>45</td>
<td>51</td>
<td>42</td>
<td>40</td>
</tr>
</tbody>
</table>
In carrageenan induced paw edema model 300 & 600 mg/kg of pet ether of BW in rats showed statistically significant (p<0.01) reduction in paw edema at all time intervals. Where as chloroform, methanol extracts showed a significant (p<0.05) reduction in all time intervals at 600 mg/kg only. The % inhibition of inflammation produced by pet ether extract of BW at dose levels of 300 & 600 mg/kg was 52 & 54 respectively at 5 hr. Standard drug indomethacin showed 82%. Chloroform and methanol at 600 mg/kg showed 44 & 40% respectively.

**Table 2: Ulcerogenic potential of BW extracts in Anti-Inflammatory model[9]**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Dose</th>
<th>Ulcer score of carrageenan induced edema</th>
</tr>
</thead>
<tbody>
<tr>
<td>Indomethacin</td>
<td>10mg/kg</td>
<td>3.60±0.74</td>
</tr>
<tr>
<td>PEBW</td>
<td>300mg/kg</td>
<td>0.80±0.49</td>
</tr>
<tr>
<td>PEBW</td>
<td>600mg/kg</td>
<td>1.40±0.40</td>
</tr>
<tr>
<td>CEBW</td>
<td>300mg/kg</td>
<td>0.60±0.40</td>
</tr>
<tr>
<td>CEBW</td>
<td>600mg/kg</td>
<td>1.79±0.25</td>
</tr>
</tbody>
</table>

In carrageenan paw edema model, pet ether, chloroform extract of BW were administered for eight days at 300 & 600 mg/kg dose level. The results of the chloroform extract of BW at 600 mg/kg produced significant ulceration 1.79±0.25 (p<0.05) and there was no significant score with pet ether extract of BW (300, 600 mg/kg) and also for 300 mg/kg of chloroform extract. Where as standard indomethacin showed significant ulcer score 3.60±0.74 (p<0.001).

**Discussion**

The results in carrageenan paw edema model reveals that among the various extract of BW, pet ether extract possessed anti-inflammatory activity followed by other extracts. Among the acute models of inflammation carrageenan induced paw edema model is commonly used model to screen anti-inflammatory agents[4].

There are three inflammation phases involved in vascular response. In the first phase there is a simultaneous release of histamine and serotonin. In the second phase kinin release occur and in the third phase PG release occurs.[5] The later phase of inhibition is reported to be clinically effective anti-inflammatory agents. It can be suggested that pet ether extract of BW has certain degree of potential as an anti-inflammatory. From the results it can also suggested that methanol extract may posses anti histamine activity, as there was significant reduction in paw edema in the early phase of inflammation (in first half an hour) as stated before the early phase of inflammation is caused by histamine and serotonin.[7]
One of the major drawbacks of anti-inflammatory agents of both type steroidal and NSAID is the ability to produce gastric ulcer. In the present study therefore ulcerogenic potential was evaluated.[8] From the results it can be observed that pet ether extract of BW at 600mg/kg which give prominent activity in carrageenan induced paw edema model and did not produce any significant ulcer score although indomethacin used as standard drug produce very significant ulceration.

References