ABSTRACT

The chemical diversity within the natural products is an immensely rich source of new pharmaceuticals, cosmetics and other economically important chemicals. Presently, numerous pure compounds are being isolated from plant sources having very promising therapeutic profiles. Now, the synthetic drugs are gradually sidelined by phytomedicines due to their adverse effects and the inability to provide complete cure in certain diseases and disorders in a cost effective manner. The present study was done to evaluate the therapeutic potential of the plant leaves of *Abies webbiana*. The anti-inflammatory activity of benzene extract of *Abies webbiana* leaves was investigated by carrageenan-induced rat hind paw edema method using mercury displacement technique with the help of travelling microscope. The benzene extract showed significant activity in a dose dependent manner as compared with standard drug diclofenac sodium. The extract containing tannins which have significant role of free radical scavenging may have the ability to produce anti-inflammatory activity.

KEYWORDS: *Abies webbiana*, Anti-inflammatory activity, Carrageenan, Diclofenac sodium.

MATERIALS AND METHOD

Introduction: Natural drug therapy is not only popular in present time but also very much famous from the past. Presently, numerous pure compounds are being isolated from plant sources having better promising therapeutic profiles than synthetic drugs. Now-a-days herbal drug therapy is much more accepted because of their less adverse effects, relatively low cost and easy availability at all the times. Thus, WHO and UNICEF are very much interested to
use herbal medicines for the treatment of children and at primary health care level. The need for basic scientific investigations on medicinal plants using indigenous system becomes an important issue today.

The plant *Abies webbiana* Lindl. (Fam-Pinaceae) is commonly known as “Himalayan silver fir” in English, “Talishpatra” in Hindi and “Badar” in Kashmir in India. It is a tall evergreen tree up to 60 m in height and 3-10 m in girth which occurs in the Himalayan region from Kashmir to Assam at an altitude of 1000-4500 m forests and largely located in humid region with heavy rain fall and dense mist.\(^{[1,2]}\)

Traditionally, plant leaves have been used as astringent, carminative, stomachic, expectorant, antispasmodic and in rheumatism.\(^{[3]}\) The decoction of the leaves is useful in case of cough and phthisis, asthma, chronic bronchitis,\(^{[4,5]}\) catarrh of bladder\(^{[2]}\) and other pulmonary affections. The juice of the fresh leaves is used as family medicine in fever and acting as an antiperiodic. It is also prescribed in affections of the chest, during dentition and in the treatment of hoarseness.\(^{[6]}\)

The whole of the plant was shown anti-tumor activity in mice.\(^{[7]}\) The petroleum ether, benzene, chloroform, acetone and ethanol extracts of this plant were found to have mast cell stabilizing action, anti-inflammatory, analgesic, antipyretic, barbiturate hypnosis potentiating and anti-ulcerogenic activity in rats.\(^{[8]}\) The methanolic extract of this leaf showed anti-tussive effect in mice.\(^{[9]}\) The present study was focused to evaluate the anti-inflammatory activity of benzene extract of *Abies webbiana* leaves by carrageenan-induced rat hind paw edema method using mercury displacement technique with the help of travelling microscope.

**Plant materials:** *Abies webbiana* Lindl. (Fam-Pinaceae) leaves were collected freshly from Gangtok, Sikkim (India) in the month of October-November and were authenticated by Botanical survey of India, Shivpur (West Bengal). The voucher specimen (AW-1) has been preserved in the herbarium department for future references. The leaves with stalks were dried under shade. Then the leaves were separated from branches, pulverized by mechanical grinder, passed through 40-mesh sieve and stored in well closed container for future use.

**Extraction procedure:** The powdered leaves were extracted with benzene solvent in soxlet apparatus. After completion of extraction the solvent was removed by distillation and dried under vacuum drier and stored in desiccators.
Chemicals required: Diclofenac sodium (Diclomax: Torrent Pharmaceuticals Pvt. Ltd., Ahmadabad, India), Carrageenan (Sigma Chemicals, USA) and Tween 80 (S.D. Chemicals Pvt. Ltd., Mumbai, India).

Preparation of samples: Different dose (100 mg/kg and 200 mg/kg body wt.) of benzene extract of Abbies webbia leaves was suspended in 1% aqueous solution of Tween-80 immediately prior to use standard drug Diclofenac sodium (10 mg/kg body wt.) and 1% w/v carrageenan solution was made by normal saline.

Animal used: Swiss albino rats of either sex weighing between 150-180 gm were used for the experiment. The animals were placed in standard metallic cages and provided with basal metabolic diet and water ad libitum. The experiments were performed in a quiet room with an ambient temperature of 22±2°C. The animal house was illuminated from 6-18 hrs daily. The rats were used for the experiment after an acclimatization period of one week. The animals were kept under 18 hrs of fasting before the commencement of the experiment.

Method: (Carrageenan-induced rat hind paw edema volumes by the mercury displacement technique with help of travelling microscope).

Carrageenan-induced paw edema method is the simplest and most widely used model for studying the anti-inflammatory activity of new compounds. For this method, the rats were divided into four groups having 6 animals in each. The first group animals were served as control and received vehicle (distilled water) only. Second group animals were administered with standard drug diclofenac sodium (10 mg/kg body wt.) orally. The third group animals were treated with benzene extracts (100 mg/kg body wt.) while the fourth group animals were treated with benzene extracts (200 mg/kg body wt.) orally. A mark was made on the left hind paw of every animal just below the tibia-tarsal junction so that every time the paw could be dipped in the mercury column of plethysmometer up to the mark to ensure constant paw volume. After one hour of the treatment with standard drug and extracts respectively, an inflammatory edema was induced in the left hind paw and the paw volume was measured plethysmographically within 30 second of the injection. The relative increase in the paw volume was measured in control, standard and test groups with a time interval of 1 hr, 2 hr, 3 hr, 4 hr and 24 hr after carrageenan injection. The percentage increase in paw volume in animals treated with standard drug and different doses of benzene extract of Abies webbiana leaves compared with control animals at the same time-intervals. The percentage of
inflammation of paw volume in treated animals of benzene extracts was compared with the anti-inflammatory potential of the standard drug as in following Table-1 and Fig-1.

Table 1: Anti-inflammatory activity of benzene extract of *Abies webbiana* leaves by carrageenan induced rat hind paw edema method.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Dose (mg/kg)</th>
<th>Percentage of inflammation after carrageenan injection at hrs</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>1 hr</td>
</tr>
<tr>
<td>Control</td>
<td>---</td>
<td>35.45±0.57</td>
</tr>
<tr>
<td>Extract</td>
<td>100</td>
<td>32.35±2.22</td>
</tr>
<tr>
<td>Extract</td>
<td>200</td>
<td>21.43±1.27**</td>
</tr>
<tr>
<td>Standard</td>
<td>10</td>
<td>16.72±1.39**</td>
</tr>
</tbody>
</table>

Results shown as Mean ± SEM (n=6), *p˂0.01, **p˂0.001, signified as compared to control.

Fig 1: Anti-inflammatory response of benzene extract of *Abies webbiana* leaves.

**Statistical analysis:** The experimental results have been expressed as the mean ± standard error of mean (SEM) and the statistical significance were evaluated by using the student’s T-test. The p-values less than 0.05 imply significance activity.\(^{[10]}\)

**RESULTS**

The results were summarized in Table-1 and Fig-1. The results clearly indicate that the benzene extract of *Abies webbiana* leaves showed significant anti-inflammatory effect in carrageenan induced rat hind paw edema. In case of 100 mg/kg body weight dose, no significant anti-inflammatory response was observed. However, highly anti-inflammatory response was observed from the second hour to onwards in case of 200 mg/kg body weight dose of the extract as compared to the standard drug diclofenac sodium.
DISCUSSION
The result demonstrates that the benzene extract of Abies webbiana leaves exhibit anti-inflammatory activity against carrageenan induced rat hind paw edema. The percentage of protection was found to be effective comparable to that of standard drug diclofenac sodium which could be related to histamine, kinin and prostaglandin inhibitory activity. The benzene extract chiefly contains tannins and saponins. It is well known that due to the significant role of tannins in free radical scavenging, which have the ability to produce anti-inflammatory activity.

CONCLUSION
The benzene extract of Abies webbiana leaves showed significant anti-inflammatory effect in the carrageenan induced rat hind paw edema method. In case of 100 mg/kg body weight dose, no significant anti-inflammatory response was observed. However, highly anti-inflammatory response was observed in case of 200 mg/kg body weight dose of the extract as compared to standard drug diclofenac sodium. This anti-inflammatory response of Abies webbiana leaves due to presence of tannins which have free radical scavenging activity.

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REFERENCES
