INTRODUCTION

Traditional medicines used through near about 60 percent of the world’s population. These are not solely used intended for major health care rural areas although also in developing countries. In developed countries modern medicines are primarily used. Use of plants as a source of medicine has been inherited and is an important component of the health care system in India. The Therapeutic efficacy of many indigenous plants, for various diseases has been described by traditional herbal medicinal practitioners. Natural products are the sources of synthetic and traditional medicine. They are still the primary health care system in some parts of the world. Several activities has been studied on the plant of Annona squamosa like antimitogenic, Antihelmintic, Scavenging, Antimicrobial, Hepatoprotective, Hepatotoxic, Antiallergic, Antispasmodial, Mollicidal. Apart from different parts of the plant have been used in traditional medicine, roots as Purgative, useful in mental depression; fruits as Haematinic, Cooling, sedative, vomiting, cough, malignant tumor and strengthening muscles; seeds as Abortifacient, useful in destroying lice in the hair; bark as atonics; leaves-crushed leaves are applied on ulcers and wounds and a leaf decoction is taken in cases of dysentery. Phytoconstituents like (−)-ent-kaur-16-en-19-oyic acid and 16a,17-dihydroxy-ent-kauran-19-oyic acid known to posses Anticancer activity, Cyclic peptide, Cyclosquamosin B, has Vasorelaxant activity, Annonaine possess Antifungal activity, which are consistent with the reported uses of the plant extracts in the indigenous system of medicine. Annona squamosa (Asquamosa) L. (Family: Annonaceae), commonly known as custard apple, Small tree 3 to 5 meters hight. Leaves are hairy when young, oblong, 8 to 15 cm long, with a petiole 1 to 1.5 cm long. Flowers occur singly in the axils of the leaves, about 2.5 cm long, pendulous, three-angled, light green to yellow. Fruit is large, slightly heart-shaped, 6 to 9 cm long, the outside with knobby polygonal tubercles. When ripe, the fruit is light yellowish-green, with a white, sweet soft, delicious and juicy flesh. A.squamosa L. is a native of West Indies, Cultivated throughout the Philippines now in India.

Keeping in mind the great medicinal value of A. squamosa L. and high content of polyphenols, flavonoids, present investigation was planned to study the analgesic activity of the Methanolic extract of seeds and compare with the standard drug Nimesulide against both eddy’s hot plate & tail flick-methods.

MATERIAL AND METHODS

Extraction

The seeds of Annona squamosa were collected from Konnagar (W.B). The plant authenticated by comparing with the herbarium voucher specimen. The seeds were air dried under shade, pulverized by a mechanical grinder and passed through a 40 mesh and then stored in airtight containers. The powdered seeds (25 g) were extracted with Methanol for 72 hr. using maceration process. This Methanolic extract was concentrated to dryness under reduced pressure and controlled temperature (50–60°C) to yield solid masses. The concentrated extracts were used for preliminary screening of phytochemicals.

Phytochemical and pharmacological evaluation of the seeds of Annona squamosa Linn.

INTRODUCTION

The aim of present study was to assess the analgesic activity of seeds of Annona squamosa Linn. The mature plant seeds of Annona squamosa were collected and authenticated. Extraction of dried seeds was carried out with methanol by maceration. Painful reaction in the experimental animals can be produced by applying unpleasant stimuli such as thermal (radiant heat as source of pain) and physical pressure (tail compression). In both Eddy’s hot plate method and tail-flick method the seed extract (250 mg/kg body weight) showed the significant analgesic activity and comparable with the standard drug Nimesulide (50mg/kg body weight).

Keywords: Annona Squamosa, Analgesic activity, Nimesulide.

ABSTRACT

The aim of present study was to assess the analgesic activity of seed extracts of Annona squamosa Linn. The mature plant seeds of Annona squamosa were collected and authenticated. Extraction of dried seeds was carried out with methanol by maceration. Painful reaction in the experimental animals can be produced by applying unpleasant stimuli such as thermal (radiant heat as source of pain) and physical pressure (tail compression). In both Eddy’s hot plate method and tail-flick method the seed extract (250 mg/kg body weight) showed the significant analgesic activity and comparable with the standard drug Nimesulide (50mg/kg body weight).

Keywords: Annona Squamosa, Analgesic activity, Nimesulide.

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Acute toxicity studies

The acute oral toxicity studies were performed to study the acute toxic effects and to determine minimum lethal dose of the drug extracts. Swiss albino mice of either sex weighing 18-25 gm were used for the study. The methanolic extracts were administered orally to different groups of overnight fasted mice at the doses of 1000, 2000 and 3000 mg/kg body weight.

Analgesic activity

Analgesic activity of methanolic extracts of Annona squamosa was studied by eddy’s hot plate and tail flick method.

Eddy’s hot plate method

The mice were divided into three groups containing 3 mice each. Group I served as control. Group II served as standard and were given nimesulide (50 mg/kg body weight) orally. Group III (test) were treated orally with methanolic extract of 250 mg/kg body weight. The animals were individually placed on the hot plate maintained at 55°C, one hour after their respective treatments.

Hot tail-flick

Male or female albino mice weighing between 18-25 gm were fasted for 24 hours with water given ad libitum maintained at room temperature. The tail was dipped into a 55°C water bath, and the time taken for the animal to flick away the tail was recorded as the latency time. The mean latency time of five trials was recorded. The mean latency time was calculated for each group in each experiment.
temperature and was divided into 3 groups of 3 mice. Group I served as control. Group II served as standard and were given nimesulide (50 mg/kg body weight) orally. Group III (test) were treated orally with methanolic extract of 250 mg/kg body weight. Analgesic effect of the test substances was determined by the hot tail-flick method described by Sewell and Spencer (1976). One to two cm of the tail of mice was immersed in warm water kept constant at 50°C.  

RESULT AND DISCUSSION  
Phytochemical screening  
The curative properties are perhaps due to the presence of various secondary metabolites such as Alkaloids, amino acids, carbohydrate, fixed oil, steroids etc. in the methanolic extraction of the seeds of Annona squamosa Linn. The results are given in table-1.  

<table>
<thead>
<tr>
<th>Test for</th>
<th>Test / Reagent</th>
<th>Observation</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkaloids</td>
<td>Dragendorff's reagent</td>
<td>Reddish brown or orange precipitate is observed</td>
<td>Alkaloids present</td>
</tr>
<tr>
<td>Amino Acids</td>
<td>Wagner's reagent</td>
<td>Reddish brown precipitate</td>
<td></td>
</tr>
<tr>
<td>Carbohydrates</td>
<td>Ninhydrin test</td>
<td>White precipitate is observed</td>
<td>Carbohydrates present</td>
</tr>
<tr>
<td>Volatile Oils</td>
<td>Molisch's test</td>
<td>Presence of violet colour</td>
<td></td>
</tr>
<tr>
<td>Flavonoids</td>
<td>Saponification test</td>
<td>Purple-violet colour ring appears</td>
<td></td>
</tr>
<tr>
<td>Glycosides</td>
<td>Shinoda test</td>
<td>No globules are observed</td>
<td></td>
</tr>
<tr>
<td>Tannins</td>
<td>Borntrager's test</td>
<td>Presence of soap formation</td>
<td>Volatile oil absent</td>
</tr>
<tr>
<td>Steroids</td>
<td>Gelatin test</td>
<td>No globules are observed</td>
<td></td>
</tr>
<tr>
<td>Salkowski test</td>
<td>No precipitate is formed</td>
<td>Carbohydrates present</td>
<td></td>
</tr>
</tbody>
</table>

Acute toxicity studies  
After administration of the extracts, animals were observed continuously for the first three hours for any toxic manifestation. Thereafter, observations were made at regular intervals for 24 hrs. No such toxic effect had been seen.  

Analgesic activity by Eddy’s hot plate method  
The response time was noted as the time at which animals reacted to the pain stimulus either by paw licking or jump response, whichever appeared first. The cut off time for the reaction was 25 seconds. The result depict that the methanolic extract of the seeds of Annona squamosa Linn having analgesic activity but much affective in colorless mice in comparison to standard Drug Nimesulide. The results are given in table-2.  

Analgesic activity by Hot Tail-flick  
The reaction time was the time taken by the mice to deflect their tails. The first reading is discarded and the reaction time was taken as a mean of the next two readings. The latent period of the tail-flick response was taken as the index of analgesia and was determined before and at 5, 10, 20, 30, 40, 50 and 60 min after the administration of drug. The maximum cut off time was fixed at 25 seconds. Same analgesic activity of methanolic extract of the seeds of Annona squamosa Linn has been observed in respect of standard drug Nimesulide. The results are given in table-3.  

<table>
<thead>
<tr>
<th>Group</th>
<th>Avg. Weight (mg)</th>
<th>Group characteristic</th>
<th>Basal reaction time (sec) before treatment</th>
<th>Reaction time (sec)</th>
<th>Basal reaction time after treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>75</td>
<td>Color (tail)</td>
<td>14</td>
<td>14</td>
<td>15</td>
</tr>
<tr>
<td>Standard</td>
<td>75</td>
<td>Color (body)</td>
<td>12</td>
<td>11</td>
<td>12</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Group</th>
<th>Avg. Weight (mg)</th>
<th>Group characteristic</th>
<th>Basal reaction time (sec) before treatment</th>
<th>Reaction time (sec)</th>
<th>Basal reaction time after treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>50</td>
<td>Color (tail)</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Standard</td>
<td>50</td>
<td>Color (body)</td>
<td>12</td>
<td>12</td>
<td>12</td>
</tr>
<tr>
<td>Test</td>
<td>50</td>
<td>Colorless</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
</tbody>
</table>
CONCLUSION

From the above investigation, it is quite apparent that methanolic extract of seeds of Annona squamosa Linn, posses potent analgesic effect against different stimuli. This is evidenced by significant increase in the reaction time by stimuli in different experimental models.

ACKNOWLEDGEMENTS

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REFERENCES