ANTHELMENTIC ACTIVITY OF ANNONA SQUAMOSA LINN LEAF EXTRACTS

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INTRODUCTION
Anthelmintics or anthelminthics are the drugs or the agents that destroy or cause the expulsion of parasitic intestinal worms. Helminthiasis is a macroparasitic disease of humans and animals in which a part of the body is infested with parasitic worms such as pinworm, roundworm, or tapeworm. It can have immunomodulatory effects on the host, with implications for any co infecting pathogens1. More than half of the population of the world suffers from infection of one or the other and majority of cattle’s suffers from worm infections. Gastrointestinal parasitism of sheep and goats is one of the leading causes of mortality, producing high economic losses2. This infection can be controlled with chemical medicinal agents but improved management is the most important infection control strategy throughout the world. Chemical control of helmenths coupled with improved management has been the important worm control strategy throughout the world. However, increasing problems of development of resistance in helminths3,4 against anthelmintics have led to the proposal of screening medicinal plants for their anthelmintic activity. The plants are known to provide a rich source of botanical anthelmintics5,6. A number of medicinal plants have been used to treat parasitic infections in man and animals7,8. Albendazole is the first drug of choice for the treatment of worm infections. It is also first reported anthelmintic which promises to have useful activity against all the types of helminth parasites menacing the domestic animals9. We have focused our attention on search of herbal remedy and selected Annona squamosa, the sugar apple that grows in tropical countries.

ABSTRACT
The aim of the present study was to investigate the anthelmintic activity of the Annona squamosa (Annonaceae), leaf extract using adult earthworm, Peritima posthuma. The hexane, ethylacetate, ethanolic extracts of the crude drug at concentrations of 100mg/ml, 200mg/ml, were tested which involve determination of paralysis time and death time. Albendazole(10mg/ml) was used as standard and it was found that the Annona reticulata leaf extracts showed dose dependent activity ethyl acetate extract at a dose of 200mg/ml has shown very high significant action.

Keywords: Anthelmintic activity, Albendazole, Earthworm, Annona squamosa.
ointment of alcoholic extract of dried leaves in white petroleum jelly is used in wound healing\(^{15}\).

**MATERIAL AND METHODS**

**Collection of Plant Materials**
The leaves of *Annona squamosa* were obtained from tirumala forest area which were authenticated by Mr. D.Ramakanth raju retire botanist.

**Preparation of extracts**
Obtained leaves were shade dried and made into coarse powder and extracted successfully using various polar grade solvents like hexane, ethylacetate, ethanol. Obtained extracts were made solvent free using rota evaporator. Extracts were stored in dessicator until further use.

**Experimental Model**
Adult earthworm *Peritima posthuma* were collected (due to its anatomical and physiological resemblance with the intestinal roundworm parasites of human being\(^{16, 17}\) from moist soil, obtained from agricultural fields nearby agricultural field. Three test groups were taken each containing six earth worms of approximately equal size (8±1 cm). Albendazole was taken as standard drug and different concentrations (10mg/ml) were prepared in normal saline containing 5% DMF\(^{18-20}\). The *Annona squamosa* leaf extracts of different concentrations were prepared by dissolving in minimum quantity of DMF and making up to the final volume with normal saline to obtain 100mg/ml, 200mg/ml. One of the groups is taken as control group which was treated with normal saline containing 5% DMF. Paralysis onset time and death time of individual worms were noted. Paralysis was said to occur when the worms do not retrieve even in normal saline. Death was concluded when the worms lost their motility followed by fading away of color of worm.

**RESULTS AND DISCUSSION**
The data in Table-1 reveals that the *Annona squamosa* Linn leaf extracts showing significant anthelmintic activity compared to the standard.

<table>
<thead>
<tr>
<th>s.no</th>
<th>Treatment</th>
<th>Dose</th>
<th>Paralysis time in mins</th>
<th>Death time</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Control</td>
<td>5% DMF in saline solution</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2</td>
<td>Albendazole</td>
<td>10mg/ml</td>
<td>2.55±0.76</td>
<td>4.37±0.66</td>
</tr>
<tr>
<td>3</td>
<td>Hexane extract</td>
<td>100mg/ml</td>
<td>25.56±0.56</td>
<td>27.62±0.14</td>
</tr>
<tr>
<td>4</td>
<td>Hexane extract</td>
<td>200mg/ml</td>
<td>21.43±0.64</td>
<td>23.11±0.26</td>
</tr>
<tr>
<td>5</td>
<td>Ethyl acetate extract</td>
<td>100mg/ml</td>
<td>4.27±0.02</td>
<td>6.43±0.07</td>
</tr>
<tr>
<td>6</td>
<td>Ethyl acetate extract</td>
<td>200mg/ml</td>
<td>2.19±0.11</td>
<td>4.11±0.09</td>
</tr>
<tr>
<td>7</td>
<td>Ethanol extract</td>
<td>100mg/ml</td>
<td>12.43±0.88</td>
<td>16.12±0.45</td>
</tr>
<tr>
<td>8</td>
<td>Ethanol extract</td>
<td>200mg/ml</td>
<td>9.16±0.66</td>
<td>12.17±0.16</td>
</tr>
</tbody>
</table>

Results are expressed as mean ± SEM from six observations; Control worms were alive up to 24 hrs of observation.
From the results we can find Annona squamosa Linn leaf extracts showed dose dependent activity, out of all three extracts ethyl acetate extract at a dose of 200mg/ml showed high significant action by killing worm in 4.11±0.09 mins. Ethanolic extract showed significant by killing the worm in 12.17±0.16 mins. Hexane extract showed moderate significant action. Standard drug albendazole showed death of worm in 4.37±0.66 mins. Ethyl acetate extract has shown equal to standard drug albendazole.

CONCLUSION
From the work we conclude that traditional usage of Annona squamosa leaf for treating worm infestations has been validated.

REFERENCES