Anthelminthic Activity of *Annona Squamosa* Linn Leaves

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**Summary**

Present study reports anthelminthic activity of various extracts obtained from the leaves of *Annona squamosa* Linn against earth worms *Phertima posthuma*. Among all the extracts tested at 20mg/ml concentration, methanolic extract showed potent anthelminthic activity when compared with the standard drug albendazole.

**Key words**: *Annona squamosa* Linn, anthelminthic, *Phertima posthuma*, albendazole.

**Introduction**

*Annona squamosa* Linn (Annonaceae) commonly known as Sugar apple is a small tropical tree, indigenous to the Amazon rain forest, growing up to 20 m tall, cultivated both in the plains and on the hills like in tropical South America, Southern Mexico, the West Indies, Bahamas, Bermuda, occasionally in Southern Florida and through out India. In Traditional System of Medicine, the leaf is used as an insecticide, in skin infections, mucosa, laxative, diarrhea, dysentery, pregnancy, antiaborticaments, for treating cancerous tumors\(^6\). The phytoconstituents isolated so far from the leaves are hydroxy ketone 10-hydroxy-16-hentriacontanone\(^7\), squamocenin, annotemoyin-2, reticulatetain-2\(^8\), benzoquinoline alkaloid samaquasine A\(^9\) and acetogenins viz., annonacin, annonacin A, annonastatin\(^10\), bullatacin, bullatacinone, and squamone\(^11\). It is reported to possess antidiabetic\(^12\), analgesic, antiinflammatory\(^13\) and larvicidal\(^14\) activities.

In the present study, our aim was to evaluate the anthelminthic potential of the different extracts of the leaves of *Annona squamosa* against adult earthworm *Phertima posthuma*.
Materials and methods

Plant material
The leaves of *Annona squamosa* Linn. was collected from and authenticated by Regional Research Institute (Ay.), Bangalore (number 2008-09/ 266). A voucher specimen was deposited in the herbarium of Department of Pharmacognosy, The Oxford College of Pharmacy, Bangalore.

Drugs and Chemicals
All the chemicals and solvents were of analytical grade and standard albendazole is of Bandy Mankind Pharm Ltd., New Delhi.

Preparation of extracts
Shade dried leaves (250g) were coarsely powdered and subjected to successive solvent extraction by a process of continuous extraction (soxhlation). The extraction was done with different solvents in their increasing order of polarity such as petroleum ether, benzene, chloroform, acetone, methanol and water. Each time the marc was dried and later extracted with other solvents. All the extract were concentrated by distilling the solvent in a rotary vacuum evaporator and evaporated to dryness. The yield was found to be 4.7, 1.24, 0.36, 2.14, 5.30 and 38.82% w/w respectively with reference to the dried material. The dried extracts were suspended in normal saline (containing 0.1% Tween 80) and used for anthelmintic study.

Preliminary phytochemical screening
The coarse powder of leaves (17g) of *Annona squamosa* was subjected to solvent extraction by soxhlation. The extraction was done with different solvents in their increasing order of polarity such as petroleum ether, benzene, chloroform, acetone, methanol and water. The extracts were concentrated and subjected to various chemical tests to detect the presence of different phytoconstituents.

Evaluation of Anthelmintic activity
The anthelmintic activity was evaluated on adult Indian earthworm *Phertima posthuma* due to its anatomical and physiological resemblance with the intestinal roundworm parasites of human beings. Seven groups consisting of five earthworms in each were released into desired formulation. Each group was treated with 0.1%Tween 80 in normal saline as vehicle. Observations were made for the time taken to paralyze and / or death of individual worms. Paralysis was said to occur when the worms do not revive even in normal saline. Death was concluded when the worms loose their motility followed with fading away of their body color.

Results and Conclusion
From the table, it is very clear that methanolic extract showed better anthelmintic activity when compared with standard drug at the same concentration. Preliminary phytochemical screening of methanol extract showed the presence of alkaloids, tannins and flavonoids.
The present study justifies the folklore claims of its potential anthelmintic property. It would be interesting to isolate the possible constituents those are responsible for anthelmintic activity.

Table 1: Anthelmintic activity of different extracts of *Annona squamosa* leaves.

<table>
<thead>
<tr>
<th>Drug tested</th>
<th>Concentration (mg/ml)</th>
<th>Paralysis time (min)</th>
<th>Death time (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vehicle [Tween 80 (0.1%) in normal saline]</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
</tr>
<tr>
<td>Albendazole</td>
<td>40</td>
<td>28±2.77</td>
<td>156±9.52</td>
</tr>
<tr>
<td>Chloroform extract</td>
<td>40</td>
<td>38±4.82</td>
<td>57±6.61</td>
</tr>
<tr>
<td>Acetone extract</td>
<td>40</td>
<td>83±9.5</td>
<td>135±11.6</td>
</tr>
<tr>
<td>Methanol extract</td>
<td>40</td>
<td>27±5.7</td>
<td>110±6.46</td>
</tr>
<tr>
<td>Aqueous extract</td>
<td>40</td>
<td>72±4.15</td>
<td>100±7.89</td>
</tr>
</tbody>
</table>

Results are expressed as mean ± SD of six determinations; Vehicle worms were alive up to 24 hrs of observation.

References