Anthelmintic Activity of Extracts of Aerial Parts of *Tephrosia spinosa* (L.f.) Pres

**Abstract**

**Purpose:** To determine the anthelmintic activity of *Tephrosia spinosa* (L.f.) Pres on Indian earth worms.  
**Methods:** Chloroform and methanol extracts of *T. spinosa* were investigated for anthelmintic activity in Indian earth worms (*Pheretima posthuma*) using albendazole (10 mg/ml) as standard reference and normal saline as control. The times to achieve paralysis and death of the worms were determined.  
**Results:** The aerial parts of *T. spinosa* (L.f.) Pres exhibited significant anthelmintic activity (*p*<0.001) when compared with albendazole.  
**Conclusion:** *Tephrosia spinosa* has paralytic effect on Indian earth worms.  

**Keywords:** *Tephrosia spinosa*, *Pheretima posthuma*, Anthelmintic activity.

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**Introduction**

Helminthiasis or infection with parasitic worm is pathogenic for human beings. Immature forms of the parasites invade human beings via the skin or gastrointestinal tract (GIT) and evolve into well differentiated adult worms that have characteristics tissue distribution. Anthelmintics are drugs that may act locally to expel worms from the GIT or systematically to eradicate adult helminths or development forms that invade organs and tissues. Most of the existing anthelmintic produces side effects such as abdominal pain, loss of appetite, nausea, vomiting, headache and diarrhoea. Chemotherapy is the only treatment and effective tool to cure and control helminths infection, as effective vaccines against helminths have not been developed so far. Indiscriminate use of synthetic anthelmintic can lead to resistance of parasites. Most diseases caused by helminths are of a chronic and debilitating in nature and could
be of value in preventing the development of resistance\(^4\).

*Tephrosia spinosa* (L.f,) Pres belongs to the family Papilionaceae\(^5\) and it is a stiffy thorny shrub, known as mullukolinji commonly found in south India on dry barren lands on the coast and island to the Hills of Coimbatore, Madurai and Tirunelveli Districts\(^6\). The phytochemical studies revealed the presence of flavanoids\(^7\)\(^-\)\(^8\). It is used in traditional system of medicine for antirheumatic, antipyretic, indigestion, antidiarrheal, anti-inflammatory, stomachic, febrifuge, anthelmintic and to control excessive thirst\(^9\). No systematic studies on anthelmintic activity have been reported on *T. spinosa*. Hence efforts have been made to establish the anthelmintic activity.

**Experimental**

**Plant Material**

The aerial parts of *T. spinosa* was collected from Madurai district in June 2009 and authenticated by Dr D Stephen who is a Taxonomist at the American College, Madurai. A voucher specimen (SRMCP/09/11) was deposited in the Department of Pharmacognosy, College of Pharmacy, SRM University, Kattankulathur for future reference. The air-dried aerial parts of plant material was ground into coarse powder using cutter mill and then stored in an air-tight container for further use.

**Preparation of Extract**

The coarsely powdered plant material was defatted with hexane using cold maceration process and further subjected to extraction with chloroform followed by methanol successively by cold maceration for five days until complete extraction was effected. It was then concentrated under reduced pressure at 50 °C and finally dried in desicators. The chloroform and methanol extracts were suspended in 1% gum acacia and used for anthelmintic activity.

**Earth Worms**

Adult Indian earth worms (*Pheretima posthuma*) collected from the local earth worm breeder in the outskirts of Madurai were used for the study.

**Anthelmintic Activity**

The anthelmintic activity was evaluated on earth worms (8 ±1cm in length) washed with normal saline to remove all the extrageneous matter as previously described\(^10\)\(^-\)\(^12\). The assay was performed on adult Indian earth worm due to its anatomical and physiological resemblance with the intestinal round worm parasite of human beings\(^13\)\(^-\)\(^15\). The worms were divided into eight groups with six worms in each group and released into appropriately labeled petri dishes with the solvent composition shown in Table 1 that were made up to 50 ml with with normal saline and then evaluated for anthelmintic activity.

**Table 1:** Treatment groups of the worms and the solvents into which they were kept

<table>
<thead>
<tr>
<th>Group</th>
<th>Worm released into</th>
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<tbody>
<tr>
<td>1*</td>
<td>50 ml of 1% gum acacia in normal saline</td>
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<tr>
<td>2**</td>
<td>50 ml albendazole 10 mg/ml</td>
</tr>
<tr>
<td>3</td>
<td>50 ml chloroform extract 10 mg in 1% gum acacia in normal saline</td>
</tr>
<tr>
<td>4</td>
<td>50 ml chloroform extract 25 mg in 1% gum acacia in normal saline</td>
</tr>
<tr>
<td>5</td>
<td>50 ml chloroform extract 50 mg in 1% gum acacia in normal saline</td>
</tr>
<tr>
<td>6</td>
<td>50 ml methanol extract, 10 mg in 1% gum acacia in normal saline</td>
</tr>
<tr>
<td>7</td>
<td>50 ml methanol extract, 25 mg in 1% gum acacia in normal saline</td>
</tr>
<tr>
<td>8</td>
<td>50 ml methanol extract, 50 mg in 1% gum acacia in normal saline</td>
</tr>
</tbody>
</table>

*control; **standard

Observations were made for the time taken for paralysis and death of individual worms to occur. For each worm, paralysis was said to have occurred when it was not able to move even in normal saline and death was concluded when it lost its motility followed with fading away of its body color\(^16\). Death was also confirmed by dipping the worm in slightly warm water and the mortality of the parasite was assumed to have
occurred when all signs of movement had ceased\(^\text{17}\). The mean paralysis time and mean lethal time of the worms for the standard and each extract were recorded.

### Statistical Analysis

Results are expressed as mean ± SEM were evaluated by one way ANOVA followed by Newman Kew’s multiple range tests. Values of P < 0.001 were considered statistically significant.

### Results and Discussion

The times for paralysis and when death of the worms occurred are provided in Table 2. Both chloroform and methanol extracts of *T. spinosa* aerial parts exhibited dose dependent and significant anthelmintic activity as compared with standard drug, abendazole. Chloroform extract required least time to causes paralysis and death of the earth worm followed when compared to the extracts from methanol suggesting either higher concentrations of the compounds producing anthelmintic activity or more compounds with the activity.

The results of this work is limited to inability to report the actual compounds responsible for the activity reported because they were not isolated or investigated. However, certain intermediate polar constituents may be responsible for anthelmintic activity than polar constituents.

### Conclusion

The chloroform and methanol extracts of *T. spinosa* have anthelminthic activity. This justifies the use of the plant in folklore remedies as an anthelmintic drug of natural origin. Further studies are required to isolate the possible phytocconstituents which may be responsible for the anthelmintic activity and to explore the possible mechanism of action.

### Table 2: Anthelmintic activity of various extracts of *Tephrosia spinosa*. (L.f.) Pres

<table>
<thead>
<tr>
<th>Groups</th>
<th>Treatment</th>
<th>Time for paralysis (min)</th>
<th>Time taken for death (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Control</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>II</td>
<td>Albendazole</td>
<td>36.19 ± 0.14</td>
<td>63.12 ± 0.31</td>
</tr>
<tr>
<td>III</td>
<td>Chloroform extract</td>
<td>48.75 ± 0.16</td>
<td>98.33 ± 0.32</td>
</tr>
<tr>
<td>IV</td>
<td>Chloroform extract</td>
<td>27.10 ± 0.21</td>
<td>44.29 ± 0.41</td>
</tr>
<tr>
<td>V</td>
<td>Chloroform extract</td>
<td>14.34 ± 0.04</td>
<td>26.43 ± 0.32</td>
</tr>
<tr>
<td>VI</td>
<td>Methanol extract</td>
<td>65.80 ± 0.17</td>
<td>113.38 ± 0.29</td>
</tr>
<tr>
<td>VII</td>
<td>Methanol extract</td>
<td>41.54 ± 0.12</td>
<td>65.57 ± 0.37</td>
</tr>
<tr>
<td>VIII</td>
<td>Methanol extract</td>
<td>21.98 ± 0.15</td>
<td>42.95 ± 0.49</td>
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</tbody>
</table>

Values are expressed as mean ± S.E.M (n=6) Control worms were alive up to 24 hours of the experiment

### References
