Original Article

In vitro anthelmintic activity of *Brassica nigra* Linn. seeds

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Abstract

Alcoholic extract from the seeds of *Brassica nigra* Linn. were investigated for their anthelmintic activity against *Pheretima posthuma* and *Ascardia galli*. Various concentrations (10-100 mg/ml) of extract were tested in bioassay, which involved the determination of time of paralysis and time of death of the worms. Alcoholic extracts exhibited significant activity at highest concentration of 100 mg/ml. Piperazine citrate (10 mg/ml) was included as standard reference and distilled as control.

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Key words: *Brassica nigra* Linn, anthelmintic activity, *Pheretima posthuma* and *Ascardia galli*

Introduction

*Brassica nigra* Linn. (Brassicaceae) is commonly known as ‘Mustard’ in English and ‘Sarson’ in Hindi. The plant is cultivated throughout India and central Europe. It is an erect, simple, mostly branched, annual herb, 50 to 60 cm tall with lower leaves distinctly stalked, lyrate-pinnatisect, largest and ovate middle leaves shorter-petioled and upper leaves oblong-linear mostly entire and all very bright green. Racemose inflorescence, fruit are long erect pods, seeds globose, 1 mm. diameter, obscurely brown and black near the hilum, delicately alveolate. The extract of *Brassica nigra* Linn was reported as antidiabetic and antihyperlipidemic effect. The leaves hot with a pleasant taste increase the bile, vermicide, good for throat complaints and seeds are bitter, hot and acrid used to cure enlargement of the spleen and dispel fever, increase the bile, remove cough, tumors, anthelmintic, increase appetite, cure skin diseases, itching and destroy external parasites is mentioned in Ayurveda. Poultice of seed are used counter irritant in several complaints of nervous system, pneumonia and an emetic in narcotic poisoning. The seeds contain alkaloid, sinapine, myrosin, sinigrin, inositol, albumins, gums and colouring matters [1]. In spite of its traditional use as anthelmintic, there are no reports on systematic and scientific study of anthelmintic activity of *Brassica nigra* in the present study; we investigate the anthelmintic activity of alcoholic extracts of seeds of *Brassica nigra* Linn.

Materials and Methods

Plant material

The seeds of *Brassica nigra* Linn. was purchased from locally were identified and authenticated by Department of Botany, Smt. U. B. Bhagat Science Mahila college, Amreli, Gujarat. The powdered seed was extracted with methanol by Soxhlet apparatus. The solvent was concentrated by evaporating methanol using a rotary evaporator.
**Table 1: Anthelmintic activity of alcoholic extract of Brassica nigra Linn.**

<table>
<thead>
<tr>
<th>Test</th>
<th>Concentration (mg/ml)</th>
<th>Time taken for paralysis (P) and death (D) of worms (mins)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>P. posthuma</td>
</tr>
<tr>
<td>Control</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Alcohol extracts</td>
<td>10</td>
<td>24.40 ± 0.4</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>17.26 ± 0.1</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>8.15 ± 0.2</td>
</tr>
<tr>
<td>Piperazine citrate</td>
<td>10</td>
<td>20.37 ± 0.3</td>
</tr>
</tbody>
</table>

Results are expressed as Mean ± SEM from six observations.

**Preparation of extracts**
The seeds of Brassica nigra Linn. were cleaned and coarsely powdered. It was then passed through the 40 mesh sieve. A weighted quantity (200 gm) of the power was subjected to continuous hot extraction in Soxhlet Apparatus exhaustively. The extract was evaporated under pressure using rotary evaporator until all solvent has been removed to give an extract sample. Percentage yield of methanol extract 4.3% w/w. The dry extracts obtained were subjected to various chemical tests to detect the presence of different phytoconstituents [2,3].

**Animals**
Indian adult earth worm (Pheretima posthuma) was collected from water logged areas of Amreli and Ascardia galli (nematode) worm were obtained from freshly slaughtered fowls. Both worm types were identified at the Department of Zoology, Smt. U. B. Bhagat Science Mahila college, Amreli, Gujarat, India.

**Drugs and chemicals used**
Piperazine citrate (Noel, Mumbai) were used as reference standards. Chemicals: Methanol (95% v/v) (Rexol Ecofuels P. Ltd. Mumbai).

**Evaluation of anthelmintic activity**
The assay was performed on Indian earthworm Pheretima posthuma due to its anatomical and physiological resemblance with intestinal roundworm parasite of human beings [4-8]. Because of easy availability, has been used widely for the initial evaluation anthelmintic compounds in vitro [9-13]. Ascardia galli worms are easily available in plenty from freshly slaughtered fowls and their use, as suitable model for screening of anthelmintic drug was advocated as earlier [14-16]. Six groups of worms were used to assess the anthelmintic properties of alcoholic extracts of Brassica nigra Linn. seeds. Groups 1 were the control worms placed in distilled water; groups 2–4 were treated with 10, 50 and 100 mg/ml of methanolic extracts Brassica nigra Linn. in distilled water; group 5 with Piperazine citrate (10mg/ml) in distilled water. Each group included six worms of each type. Observations were made for the time taken to set paralysis and death of the individual worms. Mean time for the paralysis (P) in min. was noted when no movement of any sort could be observed, except when the worm was shaken vigorously; time of death (D) in min. was recorded after ascertaining the worms neither moved when shaken vigorously nor when dipped in warm water (50°C). Piperazine citrate (10 mg/ml) was included as reference compound [17].

**Results**
Preliminary phytochemical screening of alcoholic extract revealed presence of glycosides and alkaloids. As shown in Table 1, the alcoholic extract of seeds of Brassica nigra Linn. displayed significant anthelmintic properties at higher concentration. Alcoholic extract showed anthelmintic activities in dose dependant manner giving shortest of time of paralysis (P) and death (D) with 100 mg/ml concentration for both type worms. The alcoholic extract of seeds of Brassica nigra Linn. Caused paralysis in 8.15 min and death in 27.45 min against the earthworm Pheretima posthuma. The reference drug piperazine citrate showed same at 21 min and 59 min.

Ascardia galli worms also showed sensitivity to the alcoholic extracts of Brassica nigra Linn. The alcoholic extract caused paralysis in 5 min, and death in 29 min at higher concentration of 100mg/ml. Piperazine citrate did same at 12 and 41min.
Discussion

In this study, anthelmintic assay was performed on adult Indian earthworm, *Pheretima posthuma* due to its anatomical and physiological resemblance with the intestinal roundworm parasite of human beings [5-8]. Because of easy availability, earthworms have been used widely for the initial evaluation of anthelmintic compounds *in vitro* [18-19]. *Ascardia galli* worms are easily available in plenty from freshly slaughtered fowls and their use, as a suitable model for screening of anthelmintic drug was advocated earlier [14-16]. Piperazine citrate causes flaccid paralysis of worms that resulting expulsion of worms by peristalsis. Piperazine citrate has causes death of the parasite. Therefore, it is concluded that alcoholic extracts of *Brassica nigra* Linn. Seeds have potent anthelmintic activity when compared the conventionally used drug and is equipotent to standard anthelmintic drug tested against worm species. It justifies its Ayurvedic use in curing helmintic infections.

References


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