Anthelmintic Activity of *Prosopis Cineraria* Leaves Extract on Indian Earthworm (Pheretima Posthuma)

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Abstract:
The aim of presence study was to evaluate the anthelmintic activity of aqueous, chloroform and methanolic extract of plant *Prosopis Cineraria* of family Leguminosae (Fabaceae). *Prosopis* is a genus of roughly 44 species, most of which are found in dry areas of Southwest Asia, Africa, and America, primarily from western North America to Patagonia. The plant goes by many different common names. It is referred to as Jand in Punjab and Sami, Sumri in Gujarat. Anthelmintic activity of different extracts of *Prosopis Cineraria* (Linn) leaves were evaluated against Indian earthworm. Various concentrations of the extracts were bioassay for the determination of time of paralysis (p) and time of death (d) of the worm. Piperazine citrate at a concentration of 20 mg/ml has been used as reference standard. In our present study, the crude Chloroform extract demonstrated paralysis and also caused death of worms especially at the higher concentration (50 mg/ml) compared to the other extracts and standard, piperazine citrate. This study confirmed the use of plant leaves as an anthelmintic agent.

Keyword: *Prosopis Cineraria*, Anthelmintic Activity, Piperazine citrate, Pheretima Posthuma.

Introduction:
A possible multipurpose tree species is *Prosopis Cineraria* (L.), which belongs to the Fabaceae family. The tree, also known as Jandi or Khejri locally, plays a significant role in the rural economy of the northwest Indian subcontinent. *Prosopis* is a genus of roughly 44 species, most of which are found in dry areas of Southwest Asia, Africa, and America, primarily from western North America to Patagonia.[1] The plant goes by many different common names. It is referred to as Khejri in Hindi and Sanskrit, and in Rajasthan it is known as long tree is Janti. It is referred to as Jand in Punjab and Sami, Sumri in Gujarat. Telugu translations of Tamil, Jammi, and Vanni. Its Sindi name is Kandi.[2] Given the utility of every portion of the tree, it is sometimes referred to as the "wonder tree" and "lord of the desert." It is a tiny, prickly, unevenly branching plant. a 5–10 ft. tall tree. Its bark is thick, rough, grey, and deeply fissured. P. cineraria has extremely deep roots that can extend upwards of 20 metres or more.[3] For camels, goats, and donkeys, the leaves make tasty feed. As a vegetable, the pods are consumed. The flowers can produce honey production. Sand dune stabilisation and soil enhancement are other uses for khejri. The wood is perfect for heating a home. The tree's bark has laxative and contraceptive qualities. Asthma and worms are two conditions that Khejri is known to treat.[4]

Plants and their compounds have been utilised as medicine from the beginning of time.[5] An international trend has recently changed from synthetic to herbal medicine.[6] According to the World Health Organization, between 75 and 80 percent of people worldwide either exclusively or partially utilise plant-based medications for medical purposes. The rising use and abuse of current synthetic pharmaceuticals is mostly to blame for the rise in interest in drugs derived from plants. This creates a need for the discovery and creation of novel medications to treat illnesses.[5]
**Taxonomical Classification:**
- Kingdom: Plantae
- Sub Kingdom: Phanerogames
- Division: Angiosperms
- Class: Dicotyledons
- Sub-class: Polypetalae
- Series: Calyciflorae
- Order: Rosales
- Family: Leguminosae (Fabaceae)
- Genus: Prosopis
- Species: Cineraria

**Vernacular Names**
- Hindi: Jand
- Marathi: Shami
- Telangana: Jammi
- Uttar Pradesh: Chhonkara
- Rajasthan: khejri
- Kannada: Perumbai
- Gujarati: Semi
- Tamil Nadu: vanni maram
- English: Spunge tree

**Morphological characters:**

**Leaves:** Each alternating, glabrous leaf has two pinnae; the petiole and rachis are 0.5–4 cm long. The leaflets are 7-14-jugate, oval, straight to subfalcate, without nerves (or 2-4-nerved at base, the midrib excentric), greyish when dry; stipules are foliaceous, deciduous, more or less sessile, c. 3-5 mm long and c. 2-4 mm broad, with grey-green leaves.
Flower: Creamy white flowers in pedunculate spikes are around 5–12.5 cm long, with a 1.0–2.5 cm long peduncle. Calyx is about 1-1.5 mm long, cup-shaped, and has 5 teeth, some of which are hidden. 3-4 mm long, rectangular petals with recurved tips. 10 free, quickly exerted stamens with deciduous glands at the tips of the anther’s pistil hairless.

Fruit: Pods are around 12.5–25 cm long, 5-8 mm wide, cylindric-rutulose or flat, and have a coriaceous exocarp that is brittle, a mesocarp that is pulpy, and an endocarp that is papery, thin, longitudinal, and poorly developed.

Stem: The stem is typically straight and has a deep fissured, greyish-rough bark that exfoliates in a large number of tiny flakes. The branches have short, fairly straight, somewhat compressed prickles that are glabrous and thin (3–6 mm long)

Root: Tap roots are very deep and can penetrate vertically up to 20m or more. It has a strong root system.

Seeds: A pod contain 10-15 seeds, each of which is distant, longitudinal, ovate, and 6 mm long. The faces of the tegument have an open horseshoe fissural lines, and the Testa has a medium-hard brown colour.

Microscopic Characters: [7]
Colour: Externally Brownish white or brownish green in colour
Texture: Rough, Ridged and Fissured
Oduor: Aromatic odour
Taste: Slightly Pungent
Shape: Shallow Curved
Thickness: 2mm to 5mm

Macroscopic characters: [7]
Periderm: The inner narrow zone of phellem and a few layers of phellem are found in the periderm, which is made up of outer cortical tissue.

Collapse Phloem: The periderm contains secondary phloem, which is divided between an outside zone of collapsed phloem and an interior zone of non-collapsed phloem. Sieve components are crushed into little necrost. Masses in the collapsed zone. Phloem fibres are made of gelatin. The exterior cellulose wall of gelatinous fibre is thick, and the inside mucilage is undignified.

Crystal Distribution: Crystals of calcium oxalate can be found in the phloem zone that has collapsed. Prism-shaped crystals are used.

Phytochemicals:
Leaves: methyl docosanoate, 7, 24-tirucalladien-3-one, campesterol, 20- dioate, disopropyl-9,10-dihydroxyicosane-1, tricosan, stigmasterol and actacosanol. [8,9]

Flower: Flavone derivatives such as Prosogerin B, Prosogerin A, sitosterol, spicigerinepatulitin, and patuletin glycoside. [9]

Seeds: Patuletin, patulitrin, luteolin, rutin, gallic acid, prosogerin C and prosogerin E. High amount of unsaturated fatty acids is enriched in seeds with linoleic and oleic acids. [9]

Dried pods: contain trihydroxy cinnamic acid 2-hydroxy ethyl ester, maslinic acid 3-glucoside, prophylline, 5,50 -oxybis- 1,3-benzendiol, 5,3 ,4 -trihydroxy flavanone 7-glycoside. Pods also possess total carbohydrates (45 - 55%), protein (9 - 15%) and sucrose (13.16%). Amino acids isolated from pods and leaves include Aspartic acid, Histidine, Threonine, Arginine, Serine, Glutamic acid, Alanine, Proline, Glycine, Cysteine Tyrosine, Isoleucine, Valine, Phenylalanine, Leucine, Methionine and Lysine. [10]

Whole plant: Contains methyl heptacosanoate, heneicosanoic acid, 4-hydroxy benzoic acid, methyl 4-Hydroxycinnamate, methyl 2-methoxy-5-hydroxycinnamate and O-Coumaroylglycrol. [11]

Uses:
Sheeta Jala is used to crush the seeds of Shami, Haratala, and Kadali & Syonaka ash. This paste functions well as a depilatory Agent. [12] Amati Sara, the leaves of Aralu, Tinduka, Dadima, Kutaja, and Shami are helpful. [13] Fumigation with shami leaves is effective in Arshas. [14] Conch-shell (Shanku) is rubbed with breast milk in a copper jar and fumigated with Shami leaves dipped in ghee to reduce eye irritation and pain. [15] Breast milk and ghee-smeared Shami leaves applied on fruits of Udumbara in an iron pot to relieve burning, discomfort, and redness Irritability in eye conditions. [16] Using goat’s milk to pound kantakari bark, Madhuka, and Tamra Bhasma and ghee to fumigate Shami and Amalaka leaves relieves pain swollen eye ache. [17] To prevent miscarriage, flowers and sugar are combined and taken orally. For boils and other skin irritations, P. cineraria leaf paste Blisters, such as mouth ulcers in animals and leaf infusion on open skin lesions. Leaf smoke is said to be beneficial for eye infections and problems. In a scorpion bite, the bark and leaf paste are employed. The bark prescribed for osteoarthritis. [18]
Material method:
Collection Of plant Material
Fresh leaves of Shami Plant (Prosopis Cineraria) were collected from the local area at Gandhi Nagar Shahada and authentified by Dr. Santosh K Tayade. Head, Dept. Of Botany, Arts science and Commerce college, Lonkheda, Shahada, Dist.-Nandurbar. After authentification fresh leaves of plant were collected, wash under running tap water, dried under shade for period of 8 Days and then pulverized in mechanical grinder to obtain coarse powder. The dried powder was store in airtight bottles.

Animal
Indian adult Earthworms (Pheretima Posthuma) were used to study anthelmintic activity. The earthworms were collected from moist soil at local area of Lonkhede and authentified By Head Dr. R.M. Chaudhari Dept. of zoology arts science and commerce college Lonkheda, Shahada dist. Nandurbar.

Preparation of Extract
Aqueous Extract:
The course powdered material (100 gm) of leaves was soaked in distilled water (200 ml) by simple cold maceration method for continuous 7 days and then concentration was evaporation until concentration is left and then dry.

Methanol Extract:
The course powdered material (100 gm) of leaves was soaked in methanol solution (200 ml) by simple maceration method for continuous 7 days and then concentration was evaporation until concentration is left and then dry.

Chloroform Extract:
The course powdered material (100 gm) of leaves was soaked in chloroform solution (200 ml) by simple maceration method for continuous 7 days and then concentration was evaporation until concentration is left and then dry.

Chemicals:
Saline solution
Piperazine citrate
Tween 80

Anthemlinitic Activity:
The anthemlinitic assay was carried out as per the method of Ajaiveoba et al. with necessary modification. The assay was performed on adult Indian Earthworms Pheretima Posthuma due to its anatomical and physiological resemble with the intestinal round warm parasite of human being because easy of availability. Earthworm have been used widely for initial evaluation of anthemlinitic compound. 20 ml of formulation containing different concentration of crude aqueous extract (25, 50 mg/ml) were prepared and 6 worm of same type were placed in petri plate and observe them same concentration for methanol extract (25, 50 mg/ml) were prepared. Concentration of crude chloroform extract (25, 50 mg/ml) were prepared to and 3 worms of same type were placed in it and observed them. Piperazine citrate (20 mg/ml) was used as reference standard and saline solution as control observation were made for the time take for paralysis was noted when no movement of any sort could observe except when the worms were shaken vigorously. Time for death of worm were noted after ascertaining that worms neither moved when shaken vigorously nor when dipped in saline solution all the result were expressed as a mean + - SEM of 3 worms in each group.[19]
Fig. 4 Aqueous extract 25 mg/ml

Fig. 5 Aqueous Extract 50 mg/ml

Fig. 6 Chloroform Extract 25 mg/ml
Fig. 7 Chloroform Extract 50 mg/ml

Fig. 8 Methanol Extract 25 mg/ml

Fig. 9 Methanol Extract 50 mg/ml
Table 1: Anthelmintic Activity of *Prosopis Cineraria* leaves extract on Indian earthworms (Pheretima Posthuma)

<table>
<thead>
<tr>
<th>Sr. no</th>
<th>Extract</th>
<th>Concentration in mg/ml</th>
<th>Indian Earthworm (Pheretima posthuma)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Time of paralysis in min(P)</td>
</tr>
<tr>
<td>1</td>
<td>Aqueous</td>
<td>25</td>
<td>66.85 ± 1.30</td>
</tr>
<tr>
<td>2</td>
<td></td>
<td>50</td>
<td>49.95 ± 1.55</td>
</tr>
<tr>
<td>3</td>
<td>Chloroform</td>
<td>25</td>
<td>14.45 ± 1.80</td>
</tr>
<tr>
<td>4</td>
<td></td>
<td>50</td>
<td>12.05 ± 1.58</td>
</tr>
<tr>
<td>5</td>
<td>Methanol</td>
<td>25</td>
<td>18.16 ± 1.30</td>
</tr>
<tr>
<td>6</td>
<td></td>
<td>50</td>
<td>15.13 ± 1.46</td>
</tr>
<tr>
<td>7</td>
<td>Control (Normal Saline)</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>8</td>
<td>Standard (Piperazine Citrate)</td>
<td>20</td>
<td>9.48 ± 0.65</td>
</tr>
</tbody>
</table>
**Statistical Analysis:**
The data presented as mean ± SEM. The activity of extract where compared with the control. The extract showed significantly higher duration of paralysis and death value of <0.001 were considered statistically significant.

**Result And Discussion:**
The preliminary investigation of all the extract of *Prosopis Cineraria* shows presence of Tannin, alkaloids, flavonoids, carbohydrates, Terpenes, Steroids. Some of these phytoconstituents may be responsible to show a potent anthelmintic activity from the observation made higher concentration of extract produced paralytic effect much earlier and the time of death was shorter for all worms. All the extract shows anthelmintic activity but Chlorof orm extract show Anthelmintic activity in dose dependent manner giving shorter time of paralysis (P) and death (D) with 25, 50 mg/ml concentration for worms. The Methanol extract of *Prosopis Cineraria* caused paralysis time for 25 mg/ml is 14:45 min and death time 56:05 min. 50 mg/ml time for paralysis 12:05 min and death time 43:44 min respectively against worms. The reference drug piperazine citrate (20 mg/ml) showed the paralysis at 9:48 min and death time 24:15 min respectively. The experimental evidence obtained in the laboratory model could provide a rational for the traditional used for this plant as anthelmintic.

**Summary and Conclusion:**
From the above results, it is concluded that the Chloroform extract of plant *Prosopis Cineraria* show potent anthelmintic activity to standard anthelmintic drug. Further studies using in vivo model are required to carry out and established the effectiveness and pharmacological rational for the use of *Prosopis Cineraria* as an anthelmintic agent.

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**Conflict of interest:**
The author declares no conflict of interest.
Reference:
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