Pharmacognostic Studies on the Leaves of *Prosopis cineraria* (L.) Druce. Growing in South Haryana, India

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*Prosopis cineraria* (L.) Druce. (family Leguminosae, subfamily Mimosoideae), is locally known as ‘Khejari and Jandi’ to have many uses in ethnomedicine. Establishment of pharmacognostic profile of the leaves will assist in standardization and identification of samples. The present study deals with pharmacognostic examination of morphological and microscopical characters of leaves of *Prosopis cineraria* (L.) Druce including leaf constant, ash values, extractive values.

**Keyword:** *Prosopis cineraria* (L.) Druce, Pharmacognostical study, South Haryana.

1. **Introduction**

Folk medicines often coexists with formalised, education-based, and institutionalized systems of healing such as Western medicine or Great medicine system like Ayurvedic, Unani medicine, and Chinese medicines, but is distinguishable from formalized or institutionalized healing system[1]. Indian sub-continent is endowed with numerous flora and fauna, which are used for the treatment of various ailments because of their medicinal properties. In spite of spectacular advancements in modern medicine, still rural populations of India depend and rely on traditional medicines made from plants and animals[2]. There are a number of crude drugs where the plant source has not yet been scientifically identified. Hence pharmacognostic study gives the scientific information regarding the purity and quality of the plant drugs[3]. Pharmacognostical study is the preliminary step in the standardization of crude drugs[4]. The detailed pharmacognostical evaluation gives valuable information regarding the morphology, microscopical and physical characteristics of the crude drugs[5].

In the present research we have carried out pharmacognostical study on *Prosopis cineraria* (L.) Druce (family: Leguminosae, subfamily: Mimosaceae)[6] locally known as “Khejari and Jandi”[7]. Khejari is the golden tree of Indian deserts, plays a vital role in preserving the ecosystem of arid and semi-arid areas. Since all the parts of the tree are useful, it is called kalpataru. It is also known as the ‘wonder tree’ and the ‘king of desert’[8-11]. The genus *Prosopis* belongs to the family Leguminosae, subfamily Mimosaceae and comprises about 44 species. The commonly used term “mesquite” includes all leguminous trees of genus *Prosopis*[12], distributed...
mainly in dry regions of north-western India, southern India, Afghanistan, Pakistan, Arabia and Iran\textsuperscript{[13]}. It holds an important place in the rural economy in the northwest region of Indian subcontinent\textsuperscript{[14]}. The conservation of khejri trees is a religious tenet of Rajasthan's Bishnoi community. The Government of India has recently instituted the 'Amrith Devi Bishnoi National Award for Wildlife Conservation' in the memory of Amrita Devi Bishnoi, who in 1731 sacrificed her life to protect the khejari trees in Khejarali village near Jodhpur\textsuperscript{[15]}. The Khejri tree is appropriately being used as a theme for the stamp (Fig.1a) to be released on 5\textsuperscript{th} June, 1988, World Environment Day, by the Department of Posts. The Indian Government security press has released 15,00,000 number of printed stamps\textsuperscript{16}. Prosopis cineraria is prickly tree or shrub. It is evergreen or nearly so. New leaves appear before or simultaneously with the fall of the old leaves in summer. The small, yellow flowers appear from March to May after the new flush of leaves. The pods are formed soon thereafter and grow rapidly in size. The pods ripen from June to August. Growth of new foliage, flowering and fruiting occurs during the driest months March-June when other plants become leafless and dormant\textsuperscript{[17,18]}. This plant is used in pregnancy as a safeguard against miscarriage. The smoke of the leaves is good for eye troubles. The bark is used as a remedy for rheumatism, cough, common cold asthma and scorpion stings\textsuperscript{[19,20]}, A new piperidine alkaloid spicigerin, prosogerin E along with gallic acid, pautelin, luteolin and rutin\textsuperscript{21}, Prosogerin A and B were isolated from flowers\textsuperscript{22}.

2. Materials and Methods:
2.1 Collection and Authentication and Drying of the Plant Material
The leaves of plant Prosopis cineraria (L.) Druce. (Fig.1b) chosen for the present study were collected during April 2008, from Village Pali, District Mahindergarh, State Haryana, India and the collected plant sample was identified and authenticated by Dr. H.B. Singh Head, Raw Material, herbarium and museum division, NISCAIR, New Delhi (Ref. Niscair/Rhmd/Consult/-2008-09/971/02) and a sample was deposited in the department herbarium.

2.2 Pharmacognostical evaluation\textsuperscript{[4,6,14-16]}
2.2.1 Macroscopical Characters:
The following macroscopic characters for the fresh leaves were noted: Size and shape, colour, surfaces, venation, presence or absence of petiole, the apex, margin, base, lamina, texture, odour and taste.

2.2.2 Microscopical Characters:
For microscopical studies, the leaves were cut and removed from the plant and fixed in FAA (Formalin 5 ml + Acetic acid 5 ml + 70% Ethanol 90 ml). After 24 hours of fixing, the epidermal peels and transactions of leaf were taken by free hand. The sections were stained in safranin (1%), light green (1%) and mounted in DPX Mountant (a mixture of distyrene, a plasticizer, and xylene) after the customary dehydration. Some hand sections were also examined in glycerine\textsuperscript{17-19}. The presence/absence of the following was observed: epidermal cells, stomata (type and distribution) and epidermal hairs (types of trichomes and distribution). The transverse sections of the fresh leaves through the lamina and the midrib as well as a small quantity of the powdered leaves were also cleared, mounted and observed.

2.2.3 Quantitative Microscopy:
Quantitative leaf microscopy to determine palisade ratio, stomata number, stomata index, vein – islet number and veinlet termination number were carried out on epidermal strips. The leaf epidermal studies were carried out on fresh specimens. Peels were removed mechanically using forceps. They were stained in 1% safranin mounted in glycerine and made semi-permanent by ringing with DPX Mountant (a mixture of distyrene, a plasticizer, and xylene) solution. Stomatal index (SI) and stomatal number were calculated. The vein islet number, vein termination number of the leaf and palisade
ratio of lamina were determined according to the standard method\cite{20-23}.

2.3 Physico-chemical Evaluation\cite{4,6,24,25}
Parameters determined for the powdered leaves were Loss on drying, total ash, acid insoluble ash, water soluble ash, Petroleum Ether soluble extractive, alcohol soluble extractive (90% ethanol) and water-soluble extractive values.

3. Results Discussion:
3.1 Macroscopical Characters:
*Prosopis cineraria* has bipinnately compound leaves, alternate in arrangement. The leaflets were 15-18 pairs, and shaped oblong with an entire margin, apiculate apex, obtuse base, glabrous surface, reticulate venation, petiolate, and the petiole was 0.5-4 cm long. The average leaf size was 2.5 cm (length) and 1 cm (breadth). Fresh leaves are green in colour, and are odourless with a bitter taste.

3.2 Microscopical Characters:
3.2.1 Anatomy of Leaflets
The leaflet was slightly thick in the midrib region and the lamina was slightly thin. The midrib was flat on the abaxial side and a little rose on the adaxial side. It was 170 μm thick. The epidermal layer of the midrib consists of thick, semicircular, densely tannin filled cells, measuring 20 μm in thickness. There was a wide, more or less circular vascular bundle placed in the central part of the midrib. The mesophyll cells were transcurrent along the adaxial and abaxial sides of the vascular bundle (Fig. 1.2). The vascular bundles consists of 3-5 short, compact lines of xylem elements and abaxial were of phloem elements.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Upper surface</th>
<th>Lower surface</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stomatal Number</td>
<td>21.98 ± 0.67</td>
<td>24.34 ± 0.22</td>
</tr>
<tr>
<td>Palisade ratio</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Upper surface</td>
<td>9.02 ± 0.52</td>
<td></td>
</tr>
<tr>
<td>Lower surface</td>
<td>5.16 ± 0.17</td>
<td></td>
</tr>
<tr>
<td>Vein islet no.</td>
<td>14.32 ± 0.09</td>
<td></td>
</tr>
<tr>
<td>Vein termination no.</td>
<td>26.32 ± 0.12</td>
<td></td>
</tr>
<tr>
<td>Stomatal Index</td>
<td>8.46 ± 0.24</td>
<td></td>
</tr>
</tbody>
</table>

3.2.2 Lamina
The lamina was dorsiventral, and amphistomatic (stomata occur on both upper and lower side). The lamina was 130 μm thick. The epidermal cells were semicircular, thick and contain dense tannin. The stomata were located deep below the epidermis (sunked stomata). The mesophyll was differentiated into adaxial part of two layers of vertically elongated cylindrical palisade cells and abaxial portion of 4 or 5 layers of lobed spongy mesophyll cells.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Determined values*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ethanol soluble extractive</td>
<td>19±0.9 (% w/w)</td>
</tr>
<tr>
<td>Water soluble extractive</td>
<td>26±1.0 (% w/w)</td>
</tr>
<tr>
<td>Total ash</td>
<td>6.5±0.3 (% w/w)</td>
</tr>
<tr>
<td>Water soluble ash</td>
<td>4.1±0.26 (% w/w)</td>
</tr>
<tr>
<td>Acid insoluble ash</td>
<td>1.0±0.1 (% w/w)</td>
</tr>
<tr>
<td>Sulphated ash</td>
<td>2.2±0.2 (% w/w)</td>
</tr>
<tr>
<td>Loss on drying</td>
<td>11±0.7 (% w/w)</td>
</tr>
</tbody>
</table>

Mean value of three counts
3.2.3 Leaf Margin (Fig. 2.2)
The marginal part of the lamina was broadly conical with blunt end. The margin was 80 μm thick. The epidermal cells of the margin were semicircular, thick walled and do not possess tannin content. The mesophyll tissue was not differentiated into palisade and spongy types of cells.

3.3 Powder Microscopy (Fig. 3.1, 3.2, 4.1 & 4.2)
The leaf powder showed epidermal peeling, isolated vascular strands and parenchyma cells. The epidermal cells were seen, in surface view (Fig. 3.1 & 3.2). The epidermal cells were polygonal, thick walled and anticlinal walls were straight. The stoma seems to be paracytic from the guard cells. The stomata were narrowly elliptical measuring 10 X 30 μm in size. Isolated xylem elements were seen united in a bundle. The bundle consists of vessels, fibres and parenchyma cells (Fig. 4.1). Separated parenchyma cells of the mesophyll tissue were seen scattered in the powder (Fig. 4.2). The parenchyma cells possess chloroplast and starch grains.

3.3.1 Quantitative Microscopy:
The leaf constant parameters determined in the quantitative microscopy are relatively constant for plants and can be differentiate closely related species and these are not affected by age of plant, size of leaf, environmental conditions. It is relatively constant. Hence it is more significant in the evaluation of a leaf drug. In quantitative microscopy the stomatal index, vein islet number and vein termination number and palisade ratio were found and data is given in the table.1.

3.4 Physico-Chemical Evaluation:
Physico chemical parameters like Ash value shown that presence of inorganic radicals like phosphate, carbonate and silicates. The acid insoluble ash value is more so it indicates that more contamination of metal ions. Data of extractive values shown that the amount of water soluble phyto constituents is more than alcohol soluble phyto constituents in leaf. The result is given in the Table. 2.

The present study establishes macro and microscopic characteristics, physicochemical values, Quantitative microscopical standards for leaves of Prosopis cineraria. In recent years, there has been a rapid increase in the standardization of selected medicinal plants with significant potential as therapeutics due to their specific healing properties and potential actions.
In this view, pharmacognostical standardization of *Prosopis cineraria* is necessary. As the most cost effective aid in identification of a medicinal herb, microscopic characteristics have been the mainstay of classical pharmacognosy and remain a vital component of the modern monograph.

4. Conclusion
After the present investigation it can be concluded that the Pharmacognostical studies of the leaves from *Prosopis cineraria* yielded a set of qualitative and quantitative parameters or standards that can serve as an important source of information to ascertain the identity and to determine the quality and purity of the plant materials for future studies. These parameters also will serve as standard data for quality control studies of pharmaceutical preparations from the leaves of *Prosopis cineraria*.

5. Reference:
21. A. Hussain, O.P. Virmani, Dictionary of Indian medicinal plants, Central institute of medicinal
and aromatic plants, Lucknow, 1992, 1st ed, 376.