

Pharmacognostical investigation of Root and Leaf of *Lactuca sativa* L.

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ABSTRACT

Background: *Lactuca sativa* Linn. (Asteraceae) is known as *Kahu* and is widely used as the Salad because of its high nutritional value. This plant also possesses good pharmacological activity and was also useful in traditional medicine. It possesses sedative-hypnotic, anti-inflammatory, antifungal, anti-diabetic activity. So far there has been no any scientific documentation available.

Aim and Objectives: The present study was conducted on the root and leaf of *Lactuca sativa* L. to identify its pharmacognostical characteristics as per parameters of Ayurvedic Pharmacopoeia of India (API).

Materials and Methods: Plant authentication, Organoleptic, Microscopic and Powder study was performed by following standard procedures as per Ayurvedic Pharmacopoeia of India.

Results and Discussion: *Lactuca sativa* L. have simple tap root system. The fresh roots are slightly smooth to the touch, yellowish brown in color which turns brownish colour after drying. The microscopic characteristic of the root shows outer cork, followed by cortex and vascular bundle. Some of the laticiferous cells are present in the cortex region. The leaf is dorsiventral. Transverse section of leaf shows outer epidermis followed by mesophyll cells consist of upper spongy parenchyma and lower palisade cells followed by lower epidermis. Epidermal cells shows the presence of both anisocytic and anomocytic type of stomata.

Conclusion: This study would be useful in the identification and authentication of the raw drug.

KEYWORDS : *Lactuca sativa* L. Pharmacognosy, Asteraceae, Salad

INTRODUCTION

The Asteraceae family is one of the largest family of the flowering plants, comprising about 900 genera and over 13,000 species¹. *Lactuca sativa* Linn. (Asteraceae) is considered as the most important vegetable in the group of leafy vegetables. It is almost exclusively used as a fresh vegetable in salads, but some forms are also cooked. Lettuce is produced commercially in many countries worldwide and is also widely grown as a vegetable in home gardens. It is especially important as commercial crop in Asia, North and Central America, and Europe. China, U.S., Spain, Italy, India and Japan are among the world's largest producers².

Lactuca sativa is often used in traditional medicine. It is a member of the genus of *Lactuca* (lettuce). The common name in Indonesia is Selada. The characteristics of this plant are thin roots and erect stems 30-100 cm tall, branching at the top and regular spiral-shaped leaves. The leaves are colorful, green, red, yellow, gold or blue according to varieties. *L. sativa* is commonly used as hypnotic sedative. Many researchers have been conducted in order to explore the efficacy of this plant scientifically. The study include that the sap of lettuce has antifungal effect. Oil from the lettuce seeds has sedative and hypnotic properties. It was also reported that this plant has anti-diabetic effect³

Lactuca genus comprises of about 100 species, out of which 17 European, about 10 North American, 33 tropical East African and about 40 Asian species⁴. These families have 25 species which occur in India⁵. *Lactuca scariola* is found wild in western Himalayas, *Lactuca virosa* is a variety closely allied to *lactuca scariola* is a native to Europe, *Lactuca sativa* is a common garden variety. All species emits milky latex when a Stem is cut, it is called lactucarium used in 19th century as an adulterant for opium⁶. Throughout the world *Lactuca sativa* is cultivated as a salad crop. Through literature search it has been found that extract of leaves of *Lactuca sativa* is used as Analgesic, anti-inflammatory, anti-depressant and anti- coagulant properties⁷. It also displays antiaging⁸ and antioxidative property⁹. The previous study also shows that the alcoholic extract of the plant gives sedative effect¹⁰. The plants are mostly annual or perennial herbs but a few are shrubs or trees or woody climbers. Marsh plants occasionally occur in the family¹¹. Leaves usually alternate, sometimes opposite rarely whorled mostly simple, sometimes needle like or reduced to scales; margins often pinnately or palmately lobed or divided; only rarely truly

compound; exstipulate; frequently in basal rosettes; leaves usually with oil passages; some contain latex.¹². Till date no other research work has been reported on leaves and root of the plant *Lactuca sativa*. Therefore present study designed in view of detailed Pharmacognostical studies of *Lactuca sativa*.

MATERIALS AND METHODS

Collection of the sample

The fresh whole plant of *Lactuca sativa* Linn. was collected from the natural habitat i.e., the coastal area of the Rozi port, Jamnagar, Gujarat in the month of June. The collected sample were identified and authenticated with the help of different flora and databases. A verified voucher specimen was kept in the Pharmacognosy Laboratory of the Institute, Vide no. Phm/6314/2020-21 and herbarium for future reference. The sample was washed, shade dried and coarsely powdered and preserved in an air tight container for further studies. For the histological profile, the plant was preserved in a solution of FAA (70% ethyl alcohol, Glacial acetic acid, and Formalin in the ratio 90:5:5).

Organoleptic evaluation of *Lactuca sativa*

The texture, color, odor, and taste of the root, leaf and root powder as well as leaf powder were studied as per visual observation, following the standard procedure of taxonomy and verified with existing floras for authentication

Macroscopic study and Microscopic evaluation

The macroscopic observations of the mature plants, leaves and roots were noted down. For microscopic studies, some plant material preserved in 70% alcohol.

The microscopic evaluation includes the thin free handed sections taken of both – Root as well as leaf, cleared with chloral hydrate, phloroglucinol and then with hydrochloric acid. The transverse section (TS) and powder microscopic studies were conducted and microphotographs were taken using Carl Zeiss binocular microscope with an attached camera. Powder microscopic of root and leaf of *Lactuca sativa* L. also have been carried out.

Surface study and micrometric evaluation

The surface study was done to determine the type of stomata and stomatal index. Leaf epidermal studies were carried out on fresh specimens. Peels were removed mechanically. For the determination of stomatal index, the upper and lower surface/epidermal layers of clear leaf piece were peeled out separately by means of forceps and kept on the slide to mount in glycerin water.

The epidermal layer was observed under the microscope for the determination of the stomatal type and index. Stomatal index (SI) and palisade ratio were calculated as defined by Salisbury^{13,14}

$$SI = \frac{S}{E + S} \times 100$$

Where 'S' = number of stomata per unit area and 'E' = number of epidermal cells in the same area^{15,16,17} The Stomatal index (SI) of the leaf of the plant was measured by counting the number of stomata in given area of the leaf in the microscope. Stomatal index (SI), stomatal frequency, and palisade ratio have been calculated out of an average of 5 readings. Palisade ratios (PR) was calculated as the average of palisade cells (P) beneath each epidermal cell (E). Further, They were stained in 1% safranin mounted in glycerin and made semi- permanent by ringing with DPX solution.

RESULTS AND DISCUSSION

	
<p>Fig. 1 Plant (<i>Lactuca sativa</i> L.) in natural habitat</p>	<p>Fig. 2 Flower of <i>Lactuca sativa</i> L.</p>

ROOT

Organoleptic characteristics of Root: The fresh roots are slightly smooth to the touch, yellowish brown in color, sweet in taste and aromatic. The dried root powder is rough to the touch, light brown in color sweet in taste and smell as shown in Table no. 1 and Fig. 3,4.

Table 1. Organoleptic characteristics of root of *Lactuca sativa* L. (dry and fresh form)

Parameters	Fresh root	Dry root
Texture	Slightly smooth and spongy	Rough
Color	Light yellowish brown	Light brown
Taste	Sweet	Sweet
Odor	Aromatic	Sweet



Fig. 3 Fresh root



Fig. 4 Dry root

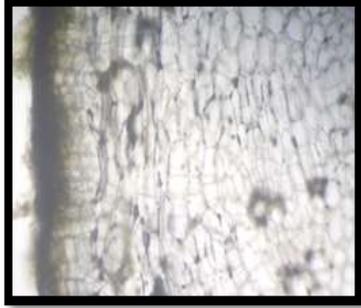
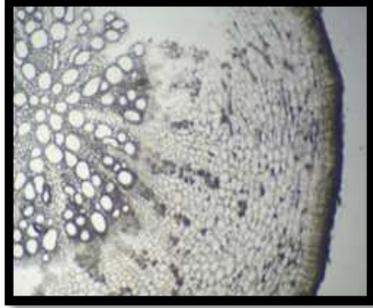
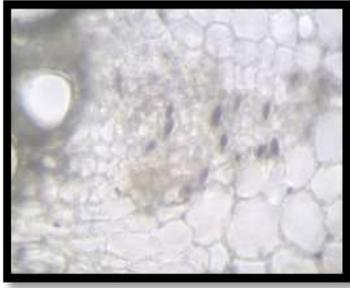
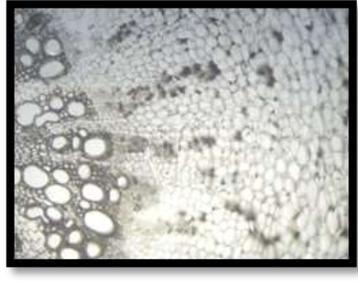
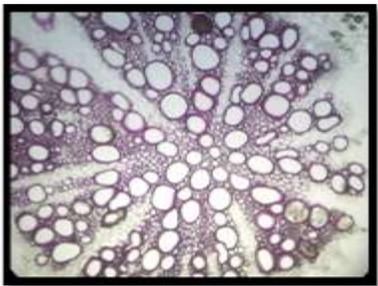
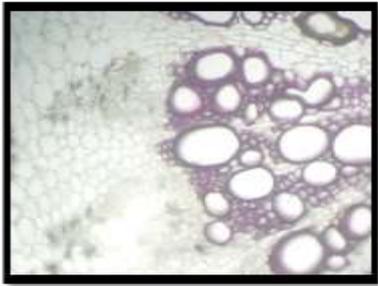
Macroscopic characteristics of the Root

The plant was identified as *Lactuca sativa* L. (Asteraceae), based on its morphological and floral characteristics. It is perennial stoloniferous herb, rooting at the nodes and forming new rosettes. The roots are simple tap root and the root stocks are semi woody, 10 cm to 1 m long and 0.5 cm broad. The roots arise from the nodal region of the plant. The young roots are yellowish brown in color, when fully matured they are light brown. The roots are aromatic, somewhat round, cylindrical and spongy with few rootlets. The fresh roots exude milky white latex when broken. The dried roots are light brown, hard, rough, semi-woody, and fibrous.

Microscopic characteristics of Root of *Lactuca sativa* L.

Transverse section: The TS taken of the root shows 8 to 9 tangentially running rows cork cells. Underneath the cortex are loosely arranged parenchyma cells with prismatic crystals and a number of laticiferous cells. Some of the cortical cells consist of brown content and are embedded with starch grains. Continuous ring of stone cells observed in the cortical zone, stone cells are with wide lumen. Discontinuous patches of lignified pericyclic fibers situated beneath the stone cells and phloem region. The vascular bundles are centrally situated and their width covers about 50% of the root. The phloem is situated above the xylem with some sieve tube and

companion cells. The medullary rays arising from the center extended upto the cortex region. It is barrel shaped multiseriate and loaded with some starch grains and prisamatic crystals. There is approximately 6 to 8 vascular bundle centrally situated. The xylem is diarch to tetrarch composed of xylem and parenchyma and xylem fibers occupying almost the entire portion of the root which is devoid of pith (Fig 5-12).

		
<p>Fig. 5 Central vascular bundle</p>	<p>Fig. 6 Cork with cortex</p>	<p>Fig. 7 Diagrammatic view</p>
		
<p>Fig. 8 Cortical cells and some brown content</p>	<p>Fig. 9 Latex content at phloem region</p>	<p>Fig. 10 Medullary rays – triseriate to multiseriate</p>
		
<p>Fig. 11 Lignified vascular bundle</p>	<p>Fig. 12 Lignified medullary rays</p>	

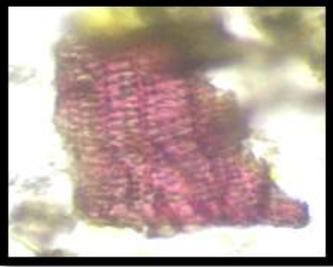
Powder microscopic of Root of *Lactuca sativa* L

Powder light brown in color, light astringent in taste and fibrous in touch. The diagnostic microscopic characteristics of the root powder shows cork in surface and tangential view and lignified and pitted parenchyma cells from cortical and vascular bundle. The cortex region contains simple parenchyma cells and simple fibers. There are also scalariform and pitted vessels of the steller region. Also starch grain in the cortex and medullary rays as well as tannin content within cell of cortical region is shown (Fig. 13-21)



Fig. 13 Powder of root of *Lactuca sativa* L.

<p>Fig. 14 Brown content</p>	<p>Fig. 15 Simple fiber</p>	<p>Fig. 16 Latex content</p>
<p>Fig. 17 Simple starch grain</p>	<p>Fig. 18 Lignified group of stone cells</p>	<p>Fig. 19 Lignified Simple fiber</p>

	
<p>Fig. 20 Scalariform vessel</p>	<p>Fig. 21 Stained fragment of pitted vessel</p>

The other family members also exhibit well develop phloem along with the laticiferous element arranged radially separated by medullary rays¹⁸.

LEAF

Organoleptic characteristic of Leaf: The fresh roots are slightly smooth to the touch, yellowish brown in color, sweet in taste and aromatic. The dried root powder is rough to the touch, light brown in color sweet in taste and smell as shown in table 2 and Fig. 22,23.

Table 2. Organoleptic characteristic of Leaf of *Lactuca sativa* L. (Fresh and dry form)

Parameters	Fresh Leaf	Dry Leaf
Texture	Smooth	Rough
Color	Green	Dark green
Taste	Salty	Sweet
Odor	Aromatic	Aromatic

	
<p>Fig. 22 Fresh leaf</p>	<p>Fig. 23 Dry leaf</p>

Morphological characteristic of Leaf

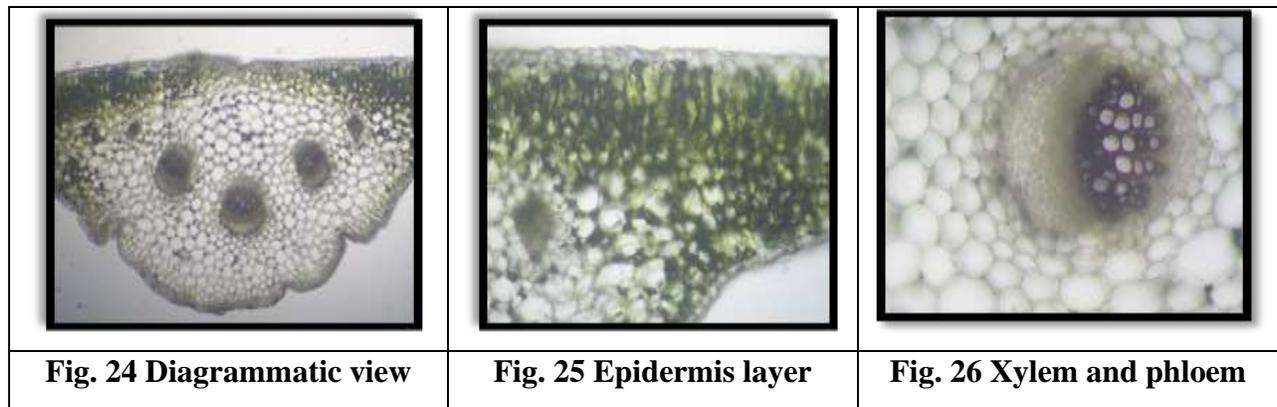
Lactuca sativa L. is a glabrous herb, with a tuft of roots at nodes. Leaves mostly radical, entire or glabrous, lower leaves obovate-oblong, pinnatifid, with rounded or very obtuse segments, spinulose on the margins with white cartilaginous teeth, cauline leaves distant, few, sessile, narrowly oblong.

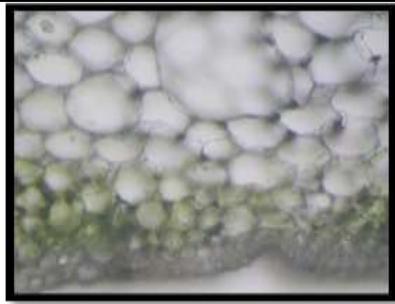
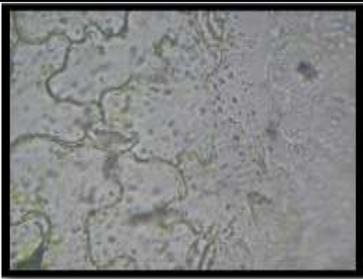
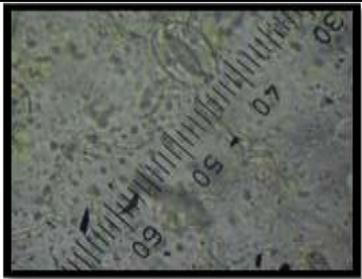
Microscopical characteristic of Leaf

Dermal Characters: Epidermal cells of adaxial epidermis have shows anisocytic and anomocytic stomata both in equal proportion. These cells with wavy walls. On abaxial surface anisocytic and anomocytic stomata are both in almost equal proportion. Epidermal cells of the abaxial epidermis are less wavy and straight walled more towards the coastal region. Epidermis is also shows non-glandular, multicellular trichomes having a single basal cell.

Transverse section: The leaf is dorsiventral, mesophyll and consist of one two three rows of upper and lower palisade cells, and 3-4 spongy parenchymous covered with thic cuticles and single layered barral shaped epidermal cells. The lower epidermis has a single layer of epidermal cells with anisocytic stomatal openings. The whole mesophyll tissue consists of chloroplast and tannin material along with some prisamatic crystals of calcium oxalate.

The transverse section through the midrib shows central vascular and lamina vascular bundle. Vascular bundles are bicolateral and consist of 2 to 5 rows of thick sclerenchyma cell. Xylems are composed of lower metaxylems and upper protoxylems. The phloem occupies both sides of the upper and lower portions of the xylem (Fig. 24-29).



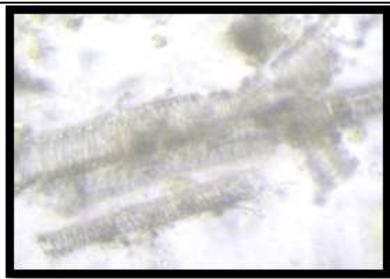
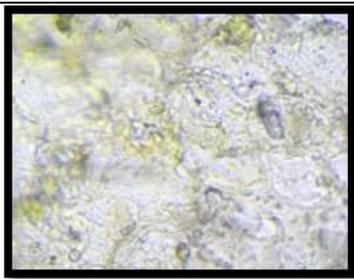
		
Fig. 27 Cholenchyma	Fig. 28 Anamocytic stomata	Fig. 29 Stomata width

Powder microscopic of Leaf of *Lactuca sativa* L.

The leaf powder is light green in color, salty in taste and spongy. The diagnostic characteristics shows the presence of simple fiber of vascular bundle. They also show pitted and spiral vessels of the stiller portion and spongy parenchyma cells and chloroplast cells from the lower mesophyll and palisade parenchyma of the upper mesophyll tissue. The lower epidermis consist of anisocytic stomata and the presence of yellowish brown content of mesophyll tissue.



Fig. 30 Powder of *Lactuca sativa* L.

		
Fig. 31 Spiral vessels	Fig. 32 Prismatic crystals	Fig. 33 Fragment of stomata along with epidermal cells

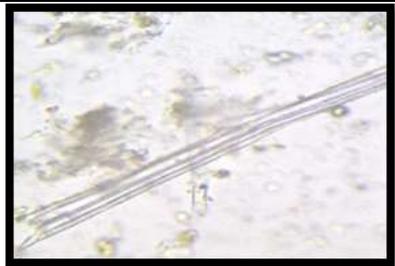
		
Fig. 34 Fiber	Fig. 35 Oil globule	Fig. 36 Latex content
		
Fig. 37 Stone cells in groups	Fig. 38 Square shaped crystals	Fig. 39 Lignified annular vessel

Table 3. Micrometric evaluation

Parameter	Size
Palisade ratio	1/3X3/epidermal cell
Stomatal index	16.67sq mm

CONCLUSION

Lactuca sativa L. is a very significant herbal drug. It is an erect, glabrous, annual herb widely grown for its crisp, highly developed radical leaves which appear before the flowering starts and exudates white milky latex from its root, leaf and stem are broken. Its root shows cork, centrally located vascular bundle, laticiferous cells, medullary rays during microscopic observation while the leaf shows epidermal cells, stomata, xylem, phloem and parenchyma which may very useful for identification and authentication of *Lactuca sativa* L. Observed parameters could be helpful to establish certain botanical standards for identification authentication and standardization of the plant. It may consider for the further research works.

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