



Pharmaceutical Standardization

Pharmacognostical evaluation of *Launaea sarmentosa* (Willd.) schultz-bip.ex Kuntze root

Yusriyya Salih, C. R. Harisha¹, Vinay J. Shukla², Rabinarayan Acharya³

Ayurvedic Physician, Wiesbaden, Germany, ¹Head, Pharamacognosy Laboratory, ²Head, Pharmaceutical Chemistry Laboratory, ³Associate Professor, Department of Dravyaguna, Institute for Post Graduate Teaching and Research in Ayurveda, Gujarat Ayurved University, Jamnagar, Gujarat, India

Abstract

Launaea sarmentosa (Willd) Schultz-Bip.ex Kuntze (Asteraceae), locally known as *Kulhafila* in the Maldives, is a creeping herb, native to tropical Indian coastlines. According to anecdotal evidence from locals in the Maldives, the roots of this plant are used as an ingredient of a popular medicinal preparation (*Hilibeys*) taken by mothers after childbirth. It is also used in various other ailments in different parts of the Maldives, as well as in India. So far, there has been no scientific documentation of this plant. The only source of information available is held by natives and traditional medical practitioners. The present study was conducted on the root of *L. sarmentosa* for its pharmacognostical and phytochemical characteristics as per Ayurvedic Pharmacopoea of India (API) parameters. The microscopic characteristics of the root show prismatic crystals, multiseriate medullary rays, laticiferous cells, and pitted parenchyma. Qualitative analyses, such as loss on drying, ash value, pH, etc., were conducted. Preliminary phytochemical screening shows the presence of alkaloids, tannin, steroids, etc.

Key words: Asteraceae, *Kulhafila*, *Launaea sarmentosa*, pharmacognosy, phytochemistry

Introduction

Launaea sarmentosa (Willd) Schultz-Bip.ex Kuntze, locally known as *Kulhafila* in the Maldives, is a prostrate, creeping, fleshy, perennial herb which is found on sandy beaches and is distributed across Mozambique, South Africa, Madagascar, Seychelles, Reunion, Mauritius, and India at an altitude range of 0 to 15 m.^[1]

The plant is used as a source of medicine in the Maldives as well as in the coastal areas of India. In the Maldives, traditional practitioners use the plant root (*Kulhafilamoo*) for various ailments. *Kulhafilamoo* (root of *L. sarmentosa*) is used mainly in a *lehya* preparation known as *Hilibeys*, which is used by mothers after child birth. It is also used for various abdominal disorders, especially due to *Vai* (*Vata*), and in urinary infections.^[2]

L. sarmentosa is also reported to possess tonic, soporific, diuretic, and aperient properties and is used as a substitute for *Taraxacum* (*Taraxacum officinale*).^[3] Latex from *L. sarmentosa* is also commonly used by fishermen to heal skin injuries caused by fish spines while fishing.^[4] Other uses in mainland India include the use of the whole plant in gout and the leaf

in rheumatism. Among the islanders of the Indian Ocean, the whole plant is used as a bath decoction to treat skin diseases and fish stings.^[5]

Through literary research, it has been found that there is no prior scientific evaluation on its pharmacognostical and phytochemical properties. The information available is mainly on its traditional use by locals living in coastal areas. In this study, the root of *L. sarmentosa* is analyzed for its pharmacognostical and phytochemical properties along with High-Performance Thin-Layer Chromatography (HPTLC) as well as tests for heavy metals.

Materials and Methods

Collection of the sample

The fresh plants of *L. sarmentosa* were collected from their natural habitat, i.e., the coastal area of the Maldives in the month of June. The collected samples were identified and authenticated with the help of different Floras^[6] and databases.^[1] A verified voucher specimen is kept in the Pharmacognosy Laboratory of IPGT and RA, Vide no: 6005/10/7/09 for future reference. The collected samples were washed, shade dried, and coarsely powdered (60 mesh) and preserved in an airtight container. For the histological profile, the plant was preserved in a solution of FAA (70% ethyl alcohol, Glacial acetic acid, and Formalin in the ration 90:5:5).^[7] Sample subjected for:

Address for correspondence: Dr. Yusriyya Salih, Fenchelring 23, 65191 Wiesbaden, Germany.
E-mail: yusriyyas@gmail.com

Pharmacognostical evaluation

Pharmacognostical evaluation was carried-out by following available standard guidelines.^[8]

Organoleptic evaluation

The texture, color, odor, and taste of the root and root powder were recorded.

Microscopic evaluation

Free-hand sections were taken, 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 a Carl Zeiss binocular microscope with an attached camera.

Histochemical analysis for starch, tannin, mucilage, lignin, and crystals were also carried out.^[9]

Physicochemical evaluation

The root was analyzed for parameters such as loss on drying, ash value, water and methanol-soluble extracts, petroleum ether extracts, and pH as per the standards of Ayurvedic Pharmacopoeia of India (API).

Preliminary phytochemical screening

Test for alkaloids, amino acids, carbohydrates, glycosides, tannin, and steroids were carried out following the standard methods as per API.

High performance thin layer chromatography

High performance thin layer chromatography (HPTLC) was carried on the saponifiable fraction of the root powder, which was subjected to petroleum ether (at 60°-80°C) in a continuous extraction apparatus (Soxhlet extractor) for 6 hours. The extract was filtered quantitatively into a tared evaporating dish and the solvent was evaporated on a water bath. The residue was dried at 105°C to constant weight. The percentage of the ether-soluble extractive value was calculated with reference to an air-dried sample of the drug. This was further subjected for the separation to obtain the unsaponifiable fraction.^[10]

Heavy metal analysis

Tests for heavy metals such as lead, arsenic, mercury, and cadmium were carried out as per the API parameters.

Results and Discussion**Organoleptic characteristics**

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 [Table 1].

Morphological characteristics of the root

The plant was identified as *L. sarmentosa* (Willd) Schultz-Bip. ex Kuntze (Asteraceae), based on its morphological [Figure 1a and b] and floral characteristics [Figure 1c]. It is a perennial stoloniferous herb, rooting at the nodes and forming new rosettes. The roots are a simple tap root and the root stocks are semi woody, 10 cm to 1 m long, and 0.5-2 cm broad. The roots arise from the nodal region of the plant. The young roots are light yellow in color, when fully matured they are light

brown. The roots are aromatic, somewhat round, cylindrical, and spongy with few rootlets [Figure 1d]. The fresh root exudes milky white latex [Figure 1e], when broken. The dried roots [Figure 1f] are light brown, hard, rough, semi-woody, and fibrous.

Microscopic characteristics**Transverse section**

The TS taken [Figure 2a] of the root shows 8 to 9 tangentially running rows of lignified cork cells. Underneath the cork are loosely arranged parenchyma cells with prismatic crystals and a number of laticiferous cells [Figure 2b]. Some of the cortical cells consist of tannin material and are embedded with starch grains. 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 [Figure 2c]. The medullary rays arising from the center extend up to the cortex region. It is barrel shaped, multi serrated, and loaded with some starch grains and prismatic crystals. There are approximately 6 to 8 vascular bundles centrally situated [Figure 2c]. The xylem is diarch to tetrarch, composed of xylem parenchyma and xylem fibers, occupying almost the entire portion of the root which is devoid of pith [Figure 2c].

The diagnostic microscopic characteristics of the root powder show cork in surface [Figure 3a] and tangential views [Figure 3b] and lignified and pitted parenchyma cells [Figure 3c] from cortical and vascular bundle. The cortex region contains simple parenchyma cells and simple fibers. There are also scalariform [Figure 3d] and pitted vessels [Figure 3e] of the stiller region. Also, starch grains in the cortex and medullary rays, as well as tannin content [Figure 3f] within cells of the cortical region are shown.

The histochemical analysis of the root powder confirms the presence of starch, tannin, mucilage, lignin, and crystals [Table 2].

Qualitative analysis

Loss on drying of the sample is directly related to moisture content and if the moisture content of the drug is high, it affects

Table 1: Organoleptic characteristics of root of *L. sarmentosa* (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

Table 2: Histochemical analysis of root of *L. sarmentosa*

Reagent	Test for	Color change	Result
Iodine	Starch	Blue	++
Ferric chloride solution	Tannin	Bluish black	++
Sudan iii	Mucilage	Red	++
Phloroglycin+HCL	Lignin	Magenta	++
Phloroglycin+HCL	Crystals	Effervescence	++

“+++”: Positive

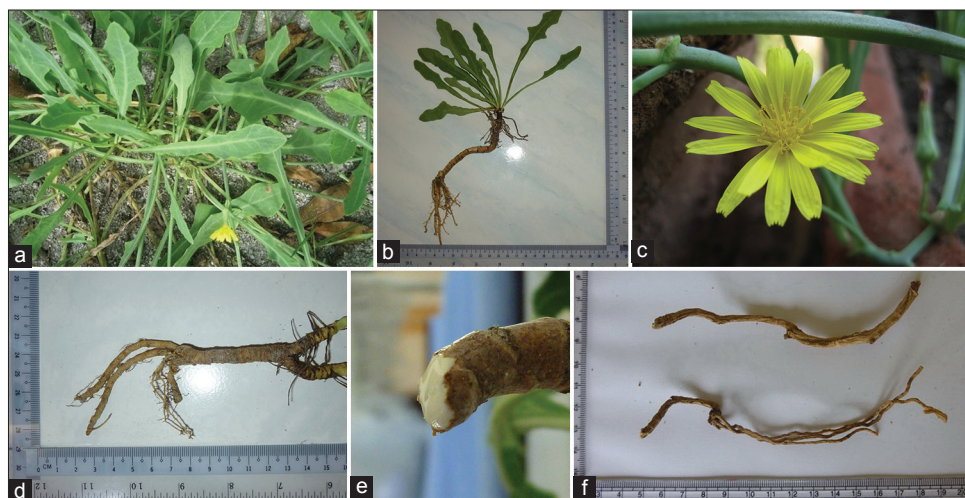


Figure 1: (a) Plant in its natural habitat (Gn.Fuvahmulah, Maldives). (b) Flowering head with ligulate florets along with stem. (c) A matured plant (d) Root exuding milky latex. (e) A matured fresh root. (f) Dried roots

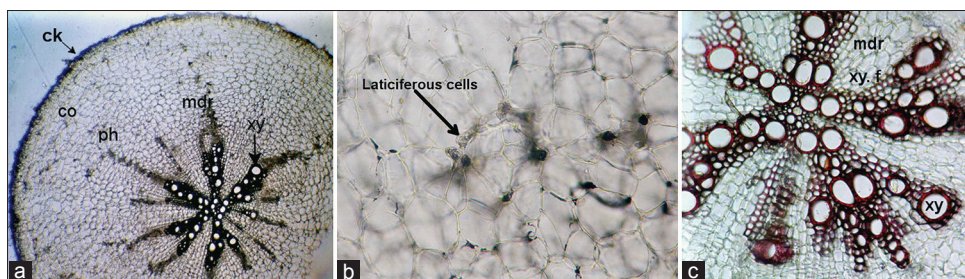


Figure 2: (a) Transverse section of the root (x3.5). (b) Laticiferous cells (x20). (c) Vascular Bundle (x10)

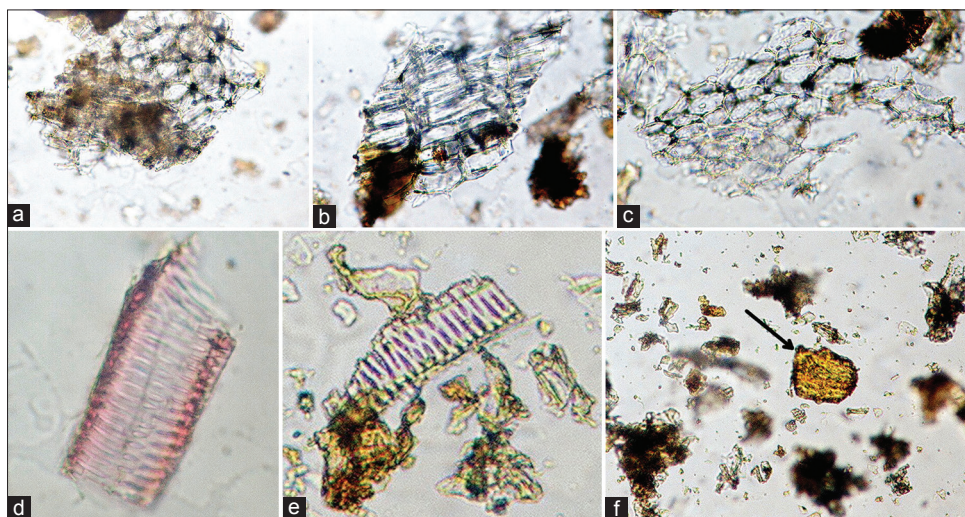


Figure 3: (a) Cork in surface view (x20). (b) Cork in tangential view (x20). (c) Parenchyma cells (x20). (d) Scalariform vessel (x20). (e) Pitted vessel (x20). (f) Tannin content (x20)

the preservation of it. Loss on drying of the sample was found to be 5.78%w/w and ash value was found to be 12.45% w/w. Ash value is indicative of the presence of inorganic and salt materials in the sample. Water- and alcohol-soluble extractive values are indicative of the bioavailability of the plant and the load of organic compounds. The water-soluble extractive value of the sample was 15.30% w/w. The alcohol-soluble extractive value was 09.78% w/w, the petroleum ether extractive value of

the drug was 2.34% w/w which indicates fixed oil content, and the pH value of the sample was found to be 6.10 which indicate that it is slightly acidic in nature [Table 3].

Preliminary phytochemical screaming

The qualitative analysis of the root powder shows the presence of alkaloids, amino acids, carbohydrates, glycosides, tannin, and steroids [Table 4].

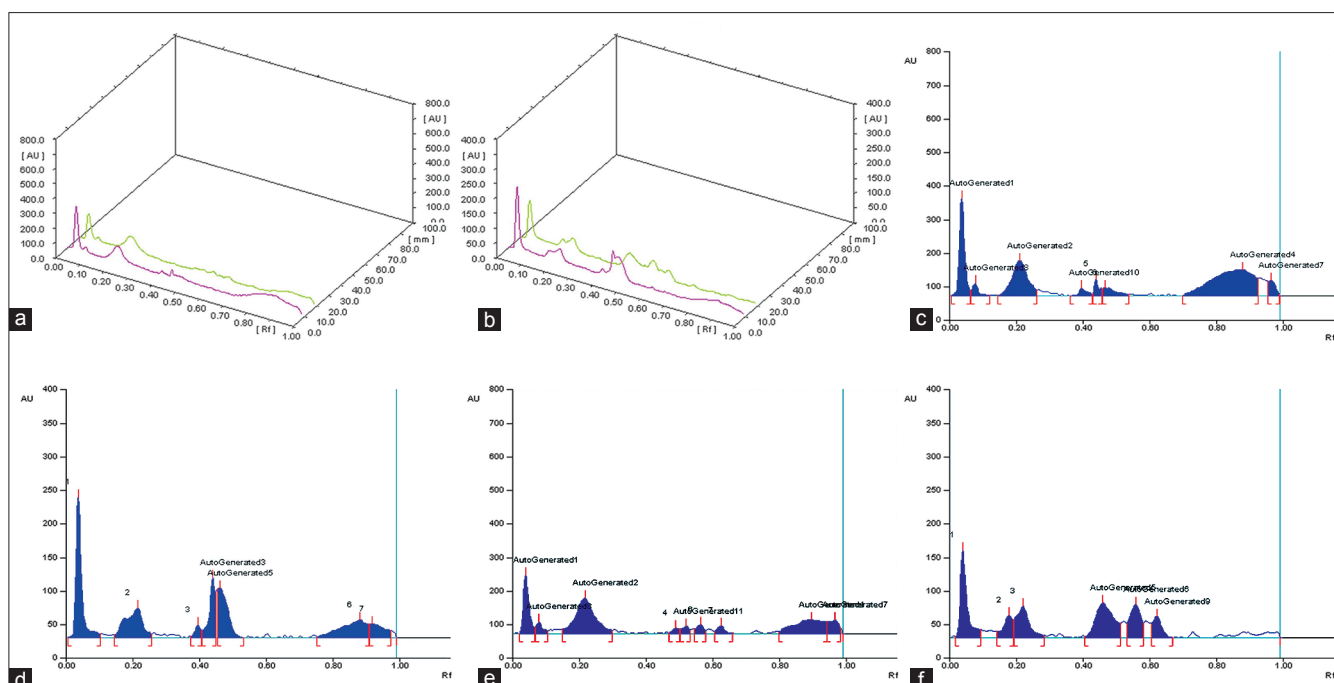


Figure 4: (a) All tracks at 254 nm. (b) All tracks at 366 nm. (c) Track 1 at 254 nm. (d) Track 1 at 366 nm. (e) Track 2 at 254 nm. (f) Track 2 at 366 nm

Table 3: Qualitative analysis of the root powder of *L. sarmentosa*

Parameter	Result
Loss on drying at 110°C	05.78% w/w
Ash value	12.45% w/w
Water-soluble extractive	15.30% w/w
Methanol-soluble extractive	09.78% w/w
Petroleum ether extractive	02.34% w/w
pH (5% w/v aqueous solution)	06.10

Table 4: Preliminary phytochemical screening of root powder of *L. sarmentosa*

Phytoconstituents	Tests (water extract)	Result
Alkaloids	Dragendorff's test	+
Amino acids	Ninhydrin test	+
Carbohydrates	Molisch's test	+
Glycosides	Fehling's test	+
Tannin	5% FeCl ₃ solution	+
Steroids	Salkowski reaction	+

“+”: Positive

Table 5: HPTLC profile for unsaponifiable fraction of root of *L. sarmentosa*

Solvent system	Track no.	Under UV			
		254 nm (Short UV)		366 nm (Long UV)	
		No. of spots	R _f value	No. of spots	R _f value
Hexane:Diethyl ether:Glacial acetic acid (7:3:0.2)	1	8	0.04, 0.08, 0.21, 0.39, 0.44, 0.47, 0.88, 0.96	7	0.04, 0.22, 0.39, 0.44, 0.46, 0.88, 0.92
	2	9	0.04, 0.08, 0.22, 0.49, 0.52, 0.56, 0.63, 0.89, 0.97	6	0.04, 0.18, 0.22, 0.46, 0.56, 0.62

Table 6: Heavy metal analysis of root powder of *L. sarmentosa*

Element	Wavelength	Instrument detection limit (ppm)	Sample results (ppm)
Lead (Pb)	220.353	0.0420	Not detected
Arsenic (As)	293.696	0.0530	Not detected
Mercury (Hg)	253.652	0.0610	Not detected
Cadmium (Cd)	228.802	0.0027	0.5530

HPTLC

The unsaponifiable fraction of the root extract of *L. sarmentosa* was subjected for chromatographic finger printing and the densitometry was labelled as Track 1 and 2 [Figure 4a, b]. Track 1 showed 8 spots at 254 nm [Figure 4c] and 7 spots at 366 nm [Figure 4d]. Track 2 showed 9 spots at 254 nm [Figure 4e] and 6 spots at 366 nm [Figure 4f]. These observations can be used as reference standards in future studies [Table 5].

Heavy metal analysis

There was no detection of lead, arsenic, and mercury,

except cadmium (0.5530 ppm). The daily value of cadmium is 0.05-0.2 micrograms/kg body.^[11] This is higher than the recommended value. The presence of heavy metal in the sample might be due to the contamination of the soil from which the plant is collected [Table 6].

Conclusion

L. sarmentosa is a perennial, prostrate, stoloniferous herb rooting at each rosette and exudate white milky latex from its root, leaf, and stem when broken. It's root showed laticiferous cells, calcium oxalate crystals, tannin content, pitted vessels, simple fibers, devoid of pith and contains alkaloids, amino acids, carbohydrates, glycosides, tannin, and steroids.

References

1. Jstor.org (homepage on the internet) JSTOR plant science; 2000-2011 ITHAKA. Available from: <http://plants.jstor.org/flora/ftca004644>. [Last accessed on 2010 Dec 27].
2. Yusriya S, Harisha CR, Shukla VJ, Acharya RN. A pharmacognostical and pharmacological evaluation of a folklore medicinal plant "Kulhafilā" *Launaea sarmentosa* (Willd) Schultz Bip.ex Kuntze). MD (Ayu) Dissertation, IPGT and RA, Gujarat Ayurved University, Jamnagar, 2011.
3. Pullaiah T. Encyclopedia of world medicinal plants. Vol. 1. Regency; 2006. p. 1217.
4. Arun AB, Beena KR, Raviraja NS, Sridhar KR. Coastal sand dunes-a neglected ecosystem (article on the internet). Available from: <http://www.ias.ac.in/currsci/jul10/articles9.htm>. [Last accessed on 2010 Oct 10].
5. Jain SK, Srivastava S. Traditional uses of some Indian plants among islanders of the Indian Ocean. Indian J Tradit Knowl 2005;4:345-57.
6. Saxena HO, Braman M. The Flora of Orissa, Vol 2. Bhubaneswar: Orissa Forest Development Corporation Ltd.; 1995. p. 946.
7. Bendre A. Practical Botany. Meerut: Rastogi Publication; 2007. p. 8-11.
8. Anonymous. The Ayurvedic Pharmacopoeia of India-Part-I, 1st ed., vol. 1. New Delhi: Department of I.S.M. and H., Ministry of Health and Family welfare, Govt. of India; 1999.
9. Kokate CK, Purohit AP, Gokhale SB. Pharmacognosy Chap. 6.1, A 1, 42nd ed. Pune: Nirali Prakashan; 2008.
10. Mangold HK. Thin layer chromatography - A Laboratory Handbook. Stahl E, editor; 2nd ed. New York: Springer - Verlag, 1969. p. 371.
11. Fda.gov (home page on the internet) U.S Food and drug Administration. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/12520187>. [Last accessed on 2011 Feb 28].

हिन्दी सारांश

पारंपारिक औषधि *Launaea sarmentosa* के मूल का फ़ार्माकोग्नोस्टिकल परीक्षण

युस्त्रिया सालिह, हरिशा सी. आर., विनय जे. शुक्ला, रबिनारायण आचार्य

Launaea sarmentosa नामक प्रसारणवती लता मालद्वीप की स्थानिक भाषा में कुल्हफ़िला नाम से जानी जाती है। भारत के तटीय इलाकों में यह उपलब्ध है। प्रसवोत्तर स्त्रियों में दी जानेवाली हिलिबीज नामक प्रसिद्ध औषधियोग में इसके मूल का उपयोग वहाँ के स्थानिक लोगों में प्रचलित है। मालद्वीप तथा भारत में उपलब्ध अन्य औषधियों में भी इस वनस्पति का उपयोग किया जाता है। लेकिन अब तक इस वनस्पति के मूल की उपयुक्तता पर वैज्ञानिक निबन्ध उपलब्ध नहीं हैं। केवल स्थानीय लोग और पारंपरिक वैद्य तक ही इसका ज्ञान सीमित है। इस विषय को ध्यान में रखकर पहली बार इसके मूल का फ़ार्माकोग्नोस्टिकल और फायटोकेमिकल परीक्षण आयुर्वेदिक फ़ार्माकोपिया के प्रमाणित स्तर पर किया गया है। फायटोकेमिकल परीक्षण में अल्कोलाइड्स टैनिन स्टेरॉइड की उपलब्धता और अनुवीक्षण यंत्र द्वारा मूल की फ़ार्माकोग्नोसी में प्रिज़मॅटिक क्रिस्टल मल्टीसिरेट मेडूलरी रेज़ की उपलब्धता प्राप्त हुई।