Original Article

Comparative pharmacognosy and phytochemical evaluation of leaf, root and stem of *Psoralea corylifolia* Linn. (*Bakuchi*)

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Abstract

Background: *Psoralea corylifolia* Linn. (*P. Corylifolia* L.), frequently familiar as *Bakuchi* in Samskrit, is an endangered and medicinally important plant. Its medicinal usage is reported in Indian pharmaceutical codex, the Chinese, British and the American Pharmacopoeia, and in different traditional systems of medicines such as Ayurveda, Unani and Siddha. However, no scientifically pharmacognosy study has been reported on leaf, root, and stem part of *P. Corylifolia* L. Classics emphasized the use of leaf, root and stem of *P. Corylifolia* L. for on the management of dental carries, diarrhea**,** dysentery, etc., in the form of local application as well as internal administration. **Aim:** The aim of this study was to evaluate comparative pharmacognosy, phytochemical studies, and physicochemical analysis of leaf, root and stem of *P. Corylifolia* L. **Materials and methods:** Studies of leaf, root, stem, and their powder for phytochemical tests, histochemical tests, psoralen chemical test, and physicochemical analysis were performed by standard methods. **Result:** All the different parts of the plant exhibit oleoresin and other cellular contents, i.e., vessels fibers, lignified pitted vessels, etc., in pharmacognosy studies. In phytochemical study; observations indicate that coumarins, steroids, and flavonoids are present in leaf, stem, and root samples. Basified alcoholic extracts of powders of all test samples showed yellowish color of fluorescence at 366 nm whereas none of the samples showed any color at 254 nm during chemical test of psoralen. **Conclusion:** Pharmacognostical study on leaf, root and stem of *Bakuchi* (*P. corylifolia* L.) contributed Certain pharmacognostical parameters i.e; oleoresin, vascular bundles, parenchyma cells with rhomboidal crystals, pericyclic fibres etc parameters that will be applicable for authentication and identification of the parts of drug. There is a need to focus on the preliminary throughput phytochemical screening of plants for their probable use in therapeutics. As no published evidences are developed on comparative pharmacognosy and preliminary physicochemical analysis of leaf, root and stem of *P. corylifolia* L. plant, the results documented in the present study may be used as a standard in subsequent studies. These observations can be of use for further research studies.

Keywords: *Bakuchi,* coumarin, pharmacognosy study, *Psoralea corylifolia* Linn., psoralen

Introduction

Bakuchi (*Psoralea corylifolia* Linn. [*P. Corylifolia L.*]) is a foremost endangered plant that has been therapeutically used to treat various manifestations for ages, which is indicated in various diseases such as *Shwitra* (vitiligo), *Kushtha* (skin diseases), *Kandu* (itching), *Jwara* (fever), *Shwasa* (asthma), and *Prameha* (diabetes).[1] Ashtanga Samgraha and Ashtanga Hridaya have mentioned it under *Shaka Varga* (classification of vegetable drugs).[2,3] Root, leaves, seed, seed oil and also the whole plant have been used for ethnomedicinal purposes.[4] The *Bakuchi Shaka* (vegetable leaves) has been indicated in diarrhea and piles in classical texts and also prescribed likewise in folklore practice. The roots are used in dental carries and leaves in diarrhea.[5,6,7] The classical indications of leaves and root of *Bakuchi* are depicted in Table 1. Phytochemical studies indicated that coumarins, flavonoids, and meroterpenes are the main components of *P. corylifolia*.[8] Recent researches carried out on *Bakuchi* have shown that it possesses all the pharmacological activities useful in skin diseases, i.e., antibacterial, anti-inflammatory, antimicrobial, antioxidant, and even anticancer.[9] The roots of *P. corylifolia* has been investigated for bioactive compounds. It was found that

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furanocoumarins psoralen and isopsoralen isolated from a petroleum ether extract were responsible for the antifeedant activity against instar *Spodoptera litura* larvae.[10] The seeds and aerial parts of *P. corylifolia* extracts with organic solvents showed activity against *Staphylococcus epidermidis* and *Morganella morganii*.[11] Raw material is having key role for its therapeutic efficacy. Adulteration may alter the efficacy of the raw material. Accordingly, proper identification of raw material is mandatory part. Published researches revealed that leaf, root, and stem of *P. Corylifolia* L. contain furanocoumarins. One can use powder of leaf, root, and stem in place of fruit as they contain furanocoumarins. These published evidences and classical references confirm therapeutic utility of leaf, root, and stem part of *P. corylifolia*. Hence, standards should be generated for proper identification of leaf, root and stem part of *P. corylifolia*.

P. corylifolia grows annually and is an erect herb. The range of height to which this plant grows is between 30 and 180 cm. It does not grow in shade and requires warm location. The soil requirement for this plant is clay, sand, and loam types. Seeds get mature in November month. The plant may grow up to 5–7 years if proper care is given.[12]

Upon review, it is revealed that leaf, root and stem had not been scientifically established as per pharmacognostical parameters although used clinically. Consequently, the present study was attempted to establish definite identification standards for leaf, root, and stem of *P. corylifolia* Linn.

Materials and methods

Whole plants of *Bakuchi* were collected from farm at Anjangaon Surji (district: Amaravati), Maharashtra, in October month at the time of its flowering and fruiting. At the time of collection, average temperature and humidity were 32°C and 74%, respectively. The details of maximum and minimum temperature and humidity during collection and drying process are depicted in Table 2. Botanical identification was done with the help of various floras and it was authenticated at the institutional pharmacognosy laboratory. Macroscopic evaluation: The leaf, root, and stem of *P. Corylifolia* L. were analyzed after making powder for the macroscopic study. Organoleptic features such as color, odor, taste, and texture of the powdered drugs were noted. Examination of the color was done under diffuse daylight. Surface characteristic, texture, and fracture characteristic were examined in samples. The material was touched to determine its softness or hardness. For odor determination, firstly the strength (none, weak distinct, and strong) and then the odor sensation (aromatic, fruity, musty, moldy, rancid, etc.) were assessed. Taste was perceived carefully by taking minute quantity of the powdered material [Table 3].

Histochemical evaluation

Thick sections of samples were subjected to histochemical tests to locate, identify classes of chemical compounds, and find starch grains, tannin, calcium, etc., treated with various reagents [Table 4].[13]

Table 1: Classical references on leaf and root of *Bakuchi*[19,20]

Reference	Used part	Indications
Charaka Samhita	Leaves	*Ama-atisara* (diarrhea), *Pravahika* (dysentery), *Arsha* (piles)
Vangasena	Root	*Krimidanta, Ruja* (dental carries and toothache)
Vaidya Manorama	Leaves (paste)	*Vrangata Rakta* (bleeding through injury)
Shodhala Nighantu	Leaves (juice)	*Shlipada* (filariasis)

Table 2: Details of temperature and humidity during collection and drying process

Particulars	At the time of collection	During drying process (5 days)	
		Minimum	Maximum
Temperature (°C)	32	24	35
Humidity (%)	74	24	96

Powder microscopy

For powder microscopy, the test materials (leaf, root, and stem of *Bakuchi*) were shade dried, and powders of leaf, root, and stem were passed through 60 no. mesh size and stored well separately in air-tight glass bottles. During drying process, 34°C average temperature and 50% humidity were noted. Total 5 days were taken for shade drying process of test materials. The drugs were individually spread on glass slides and observed under microscope at different magnifications. For the detection of lignified tissues (stone cell, sclereids, xylem vessel, etc.), the powder was stained with phloroglucinol and hydrochloric acid and to observe the starch grains, the powder was stained with iodine solution.[14]

Chemical test of psoralen

Basified alcoholic extracts of powders of test samples (i.e., leaf, root, and stem) were subjected in UV chamber for chemical test of Psoralen.[15]

Preliminary phytochemical evaluation

Methanolic extracts of leaf, root, and stem were subjected for the presence of bioactive compounds by using standard methods.[16]

Physicochemical analysis

Physicochemical analysis such as extractive values (water- and alcohol-soluble extractives), percentage of ash values, acid-insoluble ash, loss on drying (LOD) at 110°C, and pH of filtrate of 10% w/v aqueous solution (noted in Elico's digital pH meter using combined glass electrode) was carried out according to the official methods prescribed in Indian Pharmacopoeia.[17]

Results and Discussion

P. Corylifolia L. is an erect, leguminous, annual herbaceous plant that grows 60 to 100 cm tall. Macroscopy of the plant includes its branches profusely and stem is covered with white hairs. Stem measures about 15 cm × 2 cm (L × W) [Figure 1c].

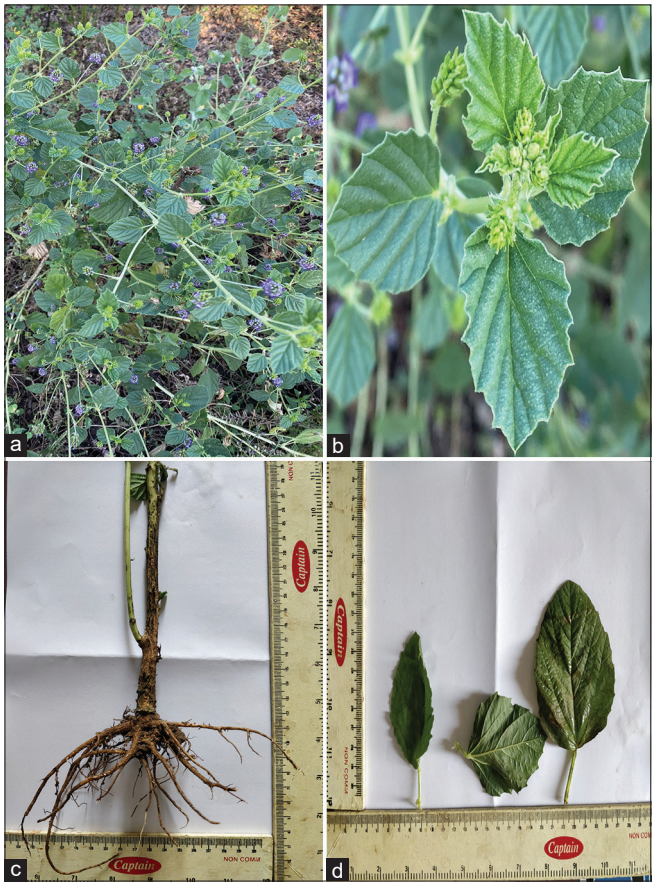

Figure 1: Morphology of whole plant of *Bakuchi*. (a) Whole plant of Bakuchi (b) leaves of Bakuchi (c) Measurement of root and stem (d) Measurement of leaf

It is grooved, rough, and green in color. Leaves are simple, measure about 8 cm × 6 cm [Figure 1d]. Leaves are rounded, hairy, and with toothed margins and both sides covered with conspicuous black glandular dots. The petioles are hairy and gland dotted. Flowers are blue in the dense axillary, solitary, 10–30 flowered racemes [Figure 1a and b]. Tap roots measure about 15 cm × 14 cm [Figure 1c]. It is observed brown in color and hard in nature and followed by the rough surface. The organoleptic characters of leaf root and stem, i.e., color, odor, taste, and touch, are tabulated in Table 3.

Microscopy of leaf shows lamina with upper and lower epidermis, transverse section (TS) of rachis with epidermis cortex and vascular bundle. Leaf through midrib shows well-defined upper and lower epidermis, upper epidermis with some trichomes, oleoresin, and lower with paracytic stomata openings. Central vascular bundle present within bundle sheath consists of vascular bundle [Figure 2]. Powder microscopy of leaf powder showed annular and pitted vessels, brown contents, crystal fibers, fragment of spongy parenchyma, fragment of stomata with epidermal cells, fragment palisade cells, oil globule, and simple fiber [Figure 3].

Diagnostic characters of root show during microscopy i.e; outer epiblema followed by a cortex with pericyclic fibers and brown content, centrally located vascular bundles with phloem, and xylem along with biserrate medullary rays [Figure 4]. Root powder showed the presence of oil globules, border pitted vessels, brown contents, group of fibers, and lignified fibers. Stained slide showed lignified cork and lignified pitted vessels [Figure 5].

Microscopy of stem show outer epidermis with wide cortex and epidermis, followed by hypodermis along with pericyclic fibers, oleoresin content cell, parenchyma cells with rhomboidal crystals, pericyclic fibers, phloem, and xylem. Centrally located pith consists of pitted parenchyma cells along with brown content, pitted parenchyma, and lignified

Table 3: Organoleptic characters of *Psoralea corylifolia* Linn. leaf, root, and stem and its powder

Organoleptic characters (*Psoralea corylifolia* Linn.)	Color	Touch	Odor	Taste
Leaf	Green	Rough	Slightly aromatic smell	Bitter-astringent
Leaf powder	Dark green	Fine fibrous	Slightly aromatic smell	Strong bitter-astringent
Root	Brown	Hard	Slightly irritating	Astringent
Root powder	Cream	Powder, fibrous	Slightly irritating	Astringent
Stem	Green	Hard	Slightly aromatic	Astringent
Stem powder	Yellowish green	Coarse, fibrous	aromatic	Bitter, astringent

Table 4: Histochemical evaluation of thick sections of leaf, root and steam of *Bakuchi*

Reagent	Observation	Characteristics	Leaf	Root	Stem
Phloroglucinol + Conc. HCl	Red	Lignified cells	+	++	++
Iodine	Blue	Starch grains	--	++	++
Phloroglucinol + Conc. HCl	Dissolved	Ca Ox - crystals	--	+	+
$Fecl_3$ solution	Dark blue	Tannin cells	+	++	++
Sudan III	Red	Oil globule	++	++	++

+ : Present, - : Absent, HCl: Hydrochloric acid

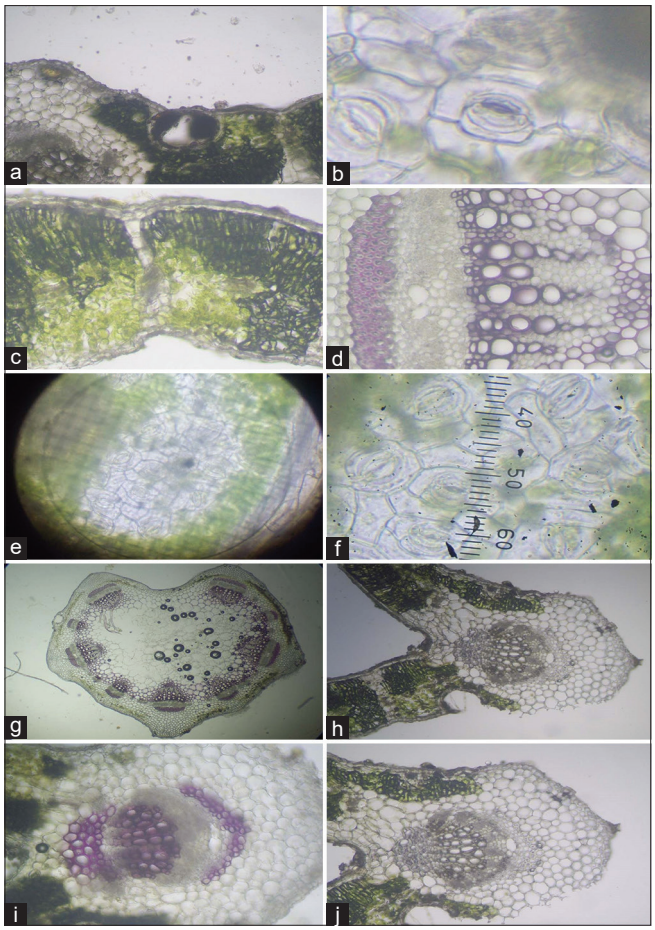

Figure 2: Transverse section of leaf (*Bakuchi*). (a) Epidermal cell with oleoresin content, (b) Epidermal cells with paracytic stomata, (c) Lamina with upper and lower epidermis, (d) Rachis with pericyclic fibers, phloem, and xylem, (e) Stomatal index, (f) Stomatal micro-measurement, (g) Transverse section of rachis, (h) Transverse section of midrib with nonlignified elements, (i) Transverse section of midrib with lignified elements, (j) Transverse section of midrib with vascular bundle

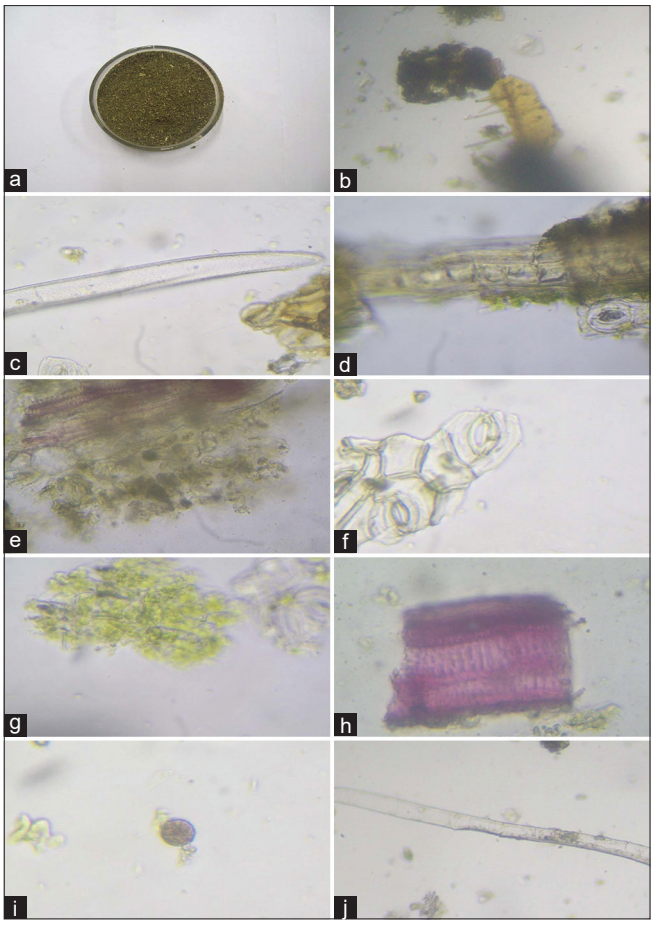

Figure 3: Powder microscopy of leaf powder (*Bakuchi*). (a) Leaf powder, (b) Brown contents, (c) Covering trichome, (d) Crystal fibers, (e) Fragment of spongy parenchyma, (f) Fragment of stomata with epidermal cells. (g) Fragment palisade cells, (h) Annular and pitted vessels, (i) Oil globule, (j) Simple fiber

elements [Figure 6]. Stem powder observed annular vessels, border pitted vessels, brown contents, group of fibers, pitted parenchymatous cells, oil globules, and trichomes. Stained slide showed lignified annular vessels, lignified group of fibers, lignified parenchymal cells, and lignified pitted vessels which are the peculiar characteristics of sample [Figure 7].

For psoralen chemical test, 2.5 g of leaf, stem, and root was taken in conical flask. Then, 50 ml of methanol was poured in it and two drops of NaOH were added. This mixture was subjected to undisturbed place for 12 h of duration. After 12 h, it was filtered through filter paper and collected liquid was taken in test tubes. Test tubes were subjected in UV chamber. None of the samples showed any color of fluorescence at 254 nm. However, all of them showed yellow-colored fluorescence at 366 nm [Table 3]. The leaf powder has shown comparatively more yellow fluorescence at 366 nm whereas stem and root powders showed dull color as compared to leaf powder at 366 nm [Table 5].

Table 5: Results of psoralen chemical test of leaf, root, and stem powder of *Bakuchi*

Samples	Color of fluorescence at 254 nm	Color of fluorescence 366 nm
Leaf powder	-	Yellowish color
Root powder	-	Yellowish white color
Stem powder	-	Yellowish orange color

The presence and absence of different bioactive compounds detected in preliminary phytochemical study are depicted in Table 6. Medicinally active constituents were observed in the plant parts samples during present investigations. Approach is done for preliminary phytochemical study from methanolic extracts of leaf, root and stem samples. These phytochemicals are known active medicinal phytoconstituents and important pharmaceuticals and are known to be of immense therapeutic importance with wide arena of therapeutic utility. In present investigation, findings indicate that coumarins, steroids, and flavonoids are more prominently present in leaf, root and stem samples. However, fatty acids were observed present in leaf and stem parts.

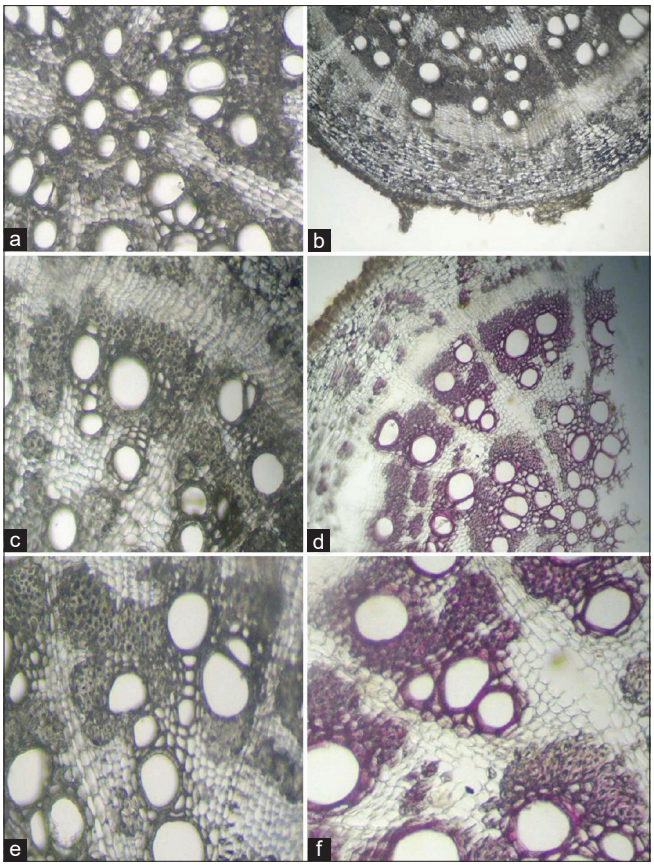

Figure 4: Transverse section of root (*Bakuchi*). (a) Centrally located vascular bundles, (b) Diagrammatic section of root with epiblema cortex and vascular bundle, (c) Transverse section with phloem and xylem, (d) Transverse section with lignified elements, pericyclic fibers, phloem, and xylem, (e) Xylem with biseriate medullary rays, (f) Lignified elements

Table 6: Presence (+) and absence (-) of active compounds in methanolic extract of leaf, root and stem of *P. corylifolia* during phytochemical screening

Plant	*Psoralea corylifolia* Linn.		
Parts (Methanol extract)	**Leaf**	**Root**	**Stem**
Compounds			
Alkaloids	+	+	-
Glycoside	-	+	-
Flavonoids	+	+	+
Tannins	+	-	-
Phenols	+	-	-
Steroids	+	+	+
Coumarins	+	+	+
Fatty acids/Lipids	+	-	+

+ : Present, - : Absent

As per the physicochemical analysis of test samples, maximum moisture content (Avg. 7.45%) was found in root sample. LOD is important for raw material which suggests its moisture regain capacity and plant material

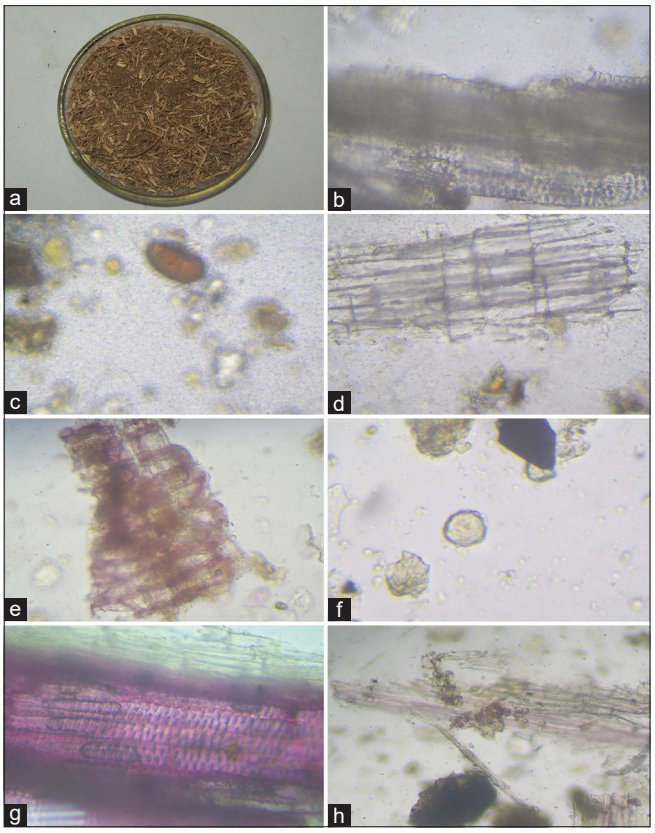

Figure 5: Powder microscopy of root powder (*Bakuchi*). (a) Root powder, (b) Border pitted vessels, (c) Brown contents, (d) Group of fibers, (e) Lignified cork cells after stain, (f) Oil globules, (g) Lignified pitted vessels, (h) Lignified fibers

which easily absorbs significant moisture and deteriorates quickly in the presence of water or more humidity. Thus, comparatively more LOD of root powder suggests need of caution for packaging, during storage, and handling. Ash value is used to determine the quality and purity of a plant material. Ash contains inorganic radicals such as phosphates, carbonates, and silicates of sodium, potassium, magnesium, calcium, etc. Major difference was not found in ash value of root, stem, and leaf samples of plant. pH was noted acidic for tested all the samples. Water-soluble and alcohol-soluble extractives determine the amounts of active constituents extracted with solvent (water/alcohol) from a given amount medicinal plants.[18] All samples were found to have comparatively more percentage of extractive value for water-soluble extractives as compared to alcohol-soluble extracts [Table 7]. The presence of maximum phytochemical functional groups in leaf than that of stem and root samples supports its classification under vegetables (*Shaka Varga*) in classics and ethnobotanic use of leaves. Thus, on the basis of preliminary phytochemical screening, it may be postulated that powder of leaf, root, and stem may also be considered for their comparative pharmacological and/or therapeutic evaluation or comparative evaluation with seed powder of *P. Corylifolia* L.

Table 7: Physicochemical analysis of leaf, root and stem of *Psoralea corylifolia* Linn. (average values)

Samples	pH	Water soluble extractives (%w/w)	Alcohol soluble extractives (%w/w)	Loss on drying (% LOD)	Total ash (%)	Acid insoluble ash (%)
Leaf powder	5.0	25.50	18.65	5.50	7.46	0.51
Root powder	6.5	28.05	20.08	7.45	7.23	1.04
Stem powder	6.0	22.04	15.08	5.60	6.12	1.01

LOD: Loss on drying

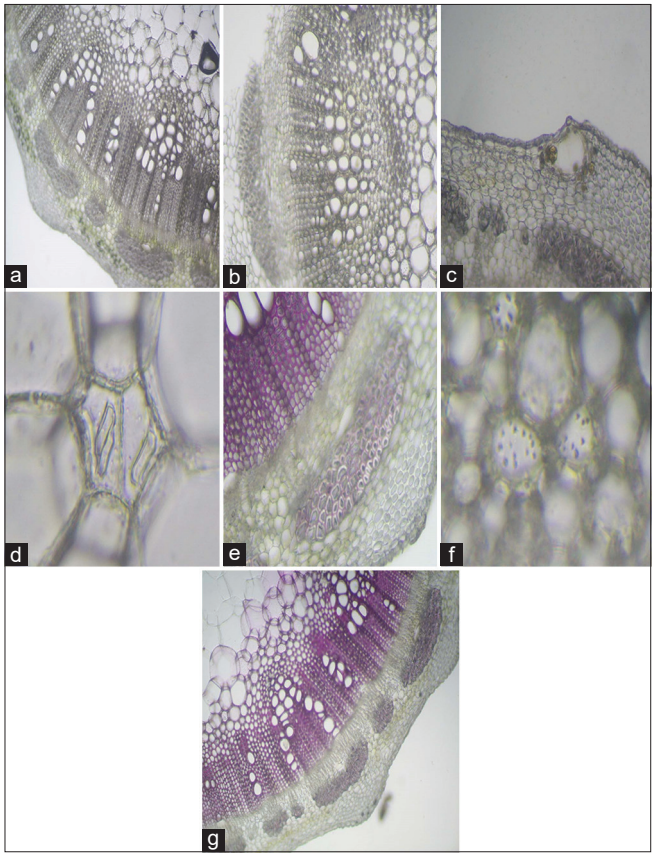

Figure 6: Transverse section of stem (*Bakuchi*). (a) Diagrammatic section of stem with epidermis cortex, vascular bundle and central pith. (b) Epidermis, hypodermis along with pericyclic fibers, (c) Oleoresin content cell. (d) Parenchyma cells with rhomboidal crystals, (e) Pericyclic fibers, phloem, and xylem. (f) Pitted parenchyma. (g) Transverse section with lignified elements

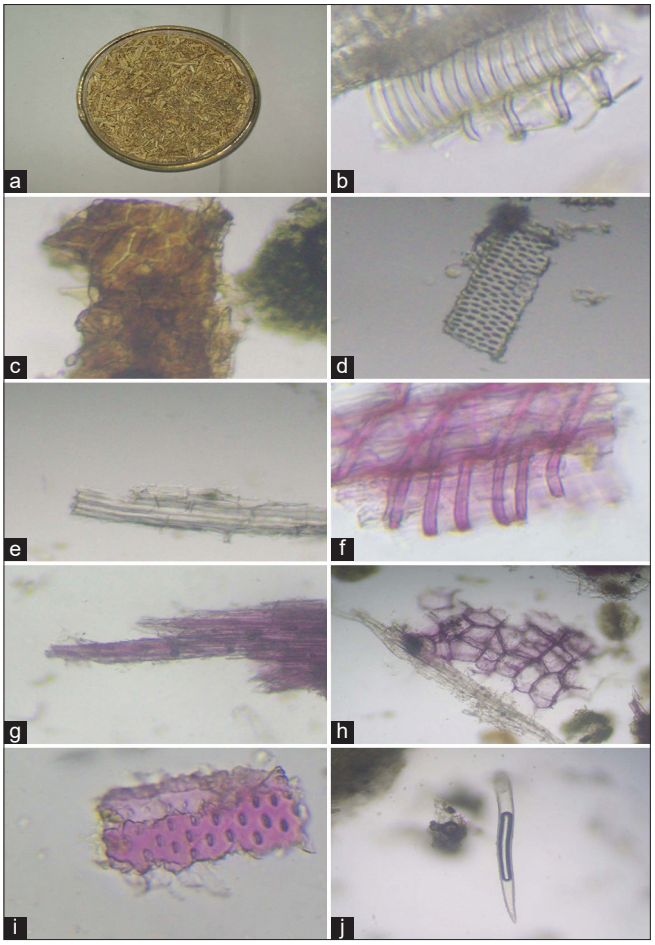

Figure 7: Powder microscopy of stem powder (*Bakuchi*). (a) Stem powder, (b) Annular vessels, (c) Brown contents, (d) Border pitted vessels, (e) Group of fibers, (f) Lignified annular vessels, (g) Lignified group of fibers, (h) Lignified parenchymal cells, (i) Lignified pitted vessels, (j) Trichomes

Conclusion

Pharmacognostical study on leaf, root and stem of *Bakuchi* (*P. Corylifolia* L.) contributed certain pharmacognostical parameters that will be applicable for authentication and identification of the parts of drug. There is a need to focus on the preliminary throughput phytochemical screening of plants for their probable use in therapeutics. Coumarins, steroids, and flavonoids are present in leaf, root and stem in preliminary phytochemical analysis. As no published evidences are developed on comparative pharmacognosy and preliminary physicochemical analysis of leaf, root and stem of *P. corylifolia* Linn. plant, the results documented in the present study may be used as a standard in subsequent studies.

Scope

Scopes are enlisted below as need for further research in view of its analytical and clinical study.

1. Comparative estimation of psoralen from leaf, root, and stem part of *P. corylifolia* Linn. by HPLC method and its application to pharmacokinetic study may be done
2. Comparative GC-MS analysis may be carried out for leaf, root, and stem part of *P. corylifolia* Linn
3. Clinical study of leaf, root, and stem powder of *P. corylifolia* Linn. may be carried out in similar diseases where seeds are indicated clinically.

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Conflicts of interest
There are no conflicts of interest.

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