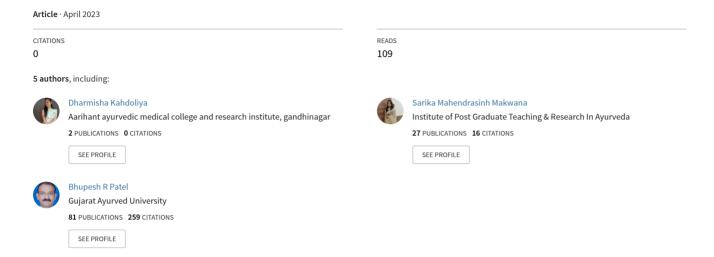
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A comparative pharmacognostical and prilimary phytochemical evaluation of *Bharangi* (Clerodendrumserratum (Linn) Kutz. and Tarkari (Clerodendrum phlomidis (Linn.) f. –root

Dharmisha H Kahdoliya^{1*}, Sarika Makwana², Sandip B Nakum³, Preeti Pandya⁴, Bhupesh Patel⁵

¹ Assistant professor, Department of Agadtantra, Arihant Ayurveda Medical College and Research Institute, Bhoyan Rathod, Gandhinagar, Gujarat, India

² PhD. (Ayu.), Department of Rasashastra, I.T.R.A, Gujarat Ayurveda University, Jamnagar, Gujarat, India
³ Assistant professor, Department of Dravyaguna, Netra Chikitsa Trust Ayurved College, Amreli, Gujarat, India
⁴ Lab Assistant, Department of Dravyaguna, I.T.R.A, Gujarat Ayurveda University, Jamnagar, Gujarat, India
⁵ Associate Professor, Department of Dravyaguna, I.T.R.A, Gujarat Ayurveda University, Jamnagar, Gujarat, India

Abstract

Ayurveda describes numerous drugs in classics. *Bharangi* is very important drug describe in various classics of Ayurveda. Which is widely used in *Shwasa*, *Kasa*, *Granthi*, *Shotha* etc diseases. Due to increased demand of *Bharangi*, it is adulterated by different plant. Thus, substitution of this plant is necessary. The drugs which have similar morphology and property are included in same family. *Acharya Bhavprakash* said that if two drugs having similar *Rasapanchaka* then one can takes as alternative of original source. Here, both drugs are used broadly in clinical practise. *Clerodendrum phlomidis* (Linn) F. used for *Meda*, *Shopha*, *Shwayathu*, *Shwas*, *Kasa*, *Pratishyay* etc. Root of both drugs have been used. Consequently, *Clerodendrum serratum* L. and *Clerodendrum phlomidis* L. both drugs belong to the same family i.e. Lamiaceae and similar *Rasadipanchaka*. Therefore, it may be *Clerodendrum phlomidis* L. used as a substitute of *Bharangi*. The study includes comparative investigations macro and microscopic characters, preliminary phytochemical analysis, physicochemical parameters and HPTLC fingerprinting of both sample drugs. Based on present phytopharmacognostical study *Clerodendrum serratum* (Linn) kutz. may be used in the scarcity of *Clerodendrum phlomidis* (Linn) F.

Keywords: lamiaceae, clerodendrum serratum (linn) kutz, clerodendrum phlomidis (linn) f, pharmacognosy, prilimary phytochemical investigation, HPTLC

Introduction

Bharangi and Tarkari both drugs are mentioned in Ayurvedic classics. Botanical source of Bharangi is Clerodendrum serratum (Linn) kutz. and Tarkari; Clerodendrum phlomidis (Linn) F. according to standard references of API and database on medicinal plants belonging to the same family Lamiaceae and genus [1]. The blue flowered shrub Bharangi is grown in lower Himalaya from Kumauni eastwards and west Bengal, Bihar up to an altitude of 1200m. It is Reported to be rare and endangered in Gujrat.[2] Traditionally root, leaves and seed used for bronchitis, Asthma, wound, Blood purifier etc.[3] A large shrub or small tree of Clerodendrum phlomidis L. is distributed throughout the India, in sub-Himalaya tracts of Bihar, Gujrat, Konkan, Kolkata etc. Bharangi is broadly used for Shwasa, Kasa, Jwara, Sotha, Vataja Granthi, Kaphaja Granthi, Raktagulma etc [4, 5, 6]. While Tarkari is useful Shwasa, Kasa, Jwara, Shwayathu, Medohara etc [7]. Both plants used individually as well as in formulations. Bharangi root used for Shwasa, Kasa etc. Tarkari patra used as vegetable, external lepa etc [8]. Root lepa also done in inflammatory condition [9]. Bharangi is highly valued and important ingredient of many popular Ayurvedic formulations like Bharangyadi leha, Kankasava, Dahmulaarista, Dashmula kwatha, etc. Tarkari as ingredient of Narayana Taila, Varunadi Kwatha, Vyaghriadi Kashaya etc. Due to increased demand of Bharangi, there is concerning reduction in availability, it is adulterated by other spurious, inferior, defective, spoiled, useless other parts of same or different plant [10]. Tarkari and Bharangi both are belonging from same family and genus and also classically having similar Rasadipanchaka. Accordingly, Tarkari is may be used in place of Bharangi. The present study includes macro and microscopic preliminary phytochemical characters. physicochemical parameters and HPTLC fingerprinting of both plants root.

Materials and Methods

Collection and authentication of plant materials

The root of *C. serratum* were collected from Lumbini Bhesaj Udyan, Sri Nrusinghnath Ayurveda College & Research Institute, Paikmal, Odisha and *C. plomidis* roots were collected from hathijana Ahmedabad, Gujarat in January 2019. Both sample were identified by local vaidya and taxonomist and confirmed by studying the morphological characters comparing them with various characters described in different floras and books [11]. Both

specimens were authenticated by pharmacognosist. Herbarium of the samples has been deposited to institute's pharmacognosy laboratory and provided with authentication number (specimen no. phm/6291/18-19 & phm/6292/18-19)

Pharmacognostical studies

The macroscopy and microscopy of both the roots were studied as per standard procedures. Thin free hand transverse section of both the roots were observed under the microscope to evaluate microscopic characters. Photographs of the section were taken with the help of Quasmo binocular compound microscope in pharmacognosy laboratory. Powders (80#) of leaf was studied for organoleptic and microscopic characters after proper mounting and staining with different reagents [12].

Physico-chemical analysis

Organoleptic examination, macro, microscopy, and physicochemical studies, viz., Ash value, water-soluble ash, water and alcohol soluble extractive value, pH value, loss on drying at 110 C as per standardized methods. [13]

Phytochemical analysis

Qualitative analysis was performed to detect primary and secondary metabolites in water and alcohol extracts of leaf. Tests for carbohydrates, protein, amino acid, steroids, glycosides, saponins, alkaloids, tannins and flavonoids, [14, 15]

HPTLC study

Methanolic root extract of *C. serratum* and *C. phlomidis* were exposed to HPTLC study. The solvent system used for the study is toluene: Formic acid (7:3:0.3) Chromatographic conditions: Application mode was Camag Linomat V, Development Chamber used was of Camag Twin trough Chamber. Precoated Silica Gel GF254 Plates were used. Chamber saturation was done for 30 min and development time was 30 min. The plate was scanned in Camag Scanner III with Deuterium lamp, Tungstan Lamp as detectors and Wincats software was used for data analysis.

Results and Discussion Morphology

Bharangi (Clerodendrum serratum L.) and Tarkari (Clerodendrum phlomidis L.) both the species had Katu, Tikta Rasa according to most of the nighantus. Colour whitish brown in Clerodendrum serratum L. and creamish yellow in Clerodendrum phlomidis L. and both perceived aromatic odour. Based on the morphology and microscopical study done by pharmacognostical methods

Microscopic characters

Transverse section of *Clerodendrum serratum* (Linn) kutz. (Fig. 1c to 1h)

The diagrammatic section of the root shows, outermost cork followed with cortex, phloem and central xylem. Details

section shows cork consisting of 10-12 layers of thin walled tangentially arranged cells having wavy radial conjoining cell walls at places. Outermost layers observed with dark brown color, while the inner layers with alteration of lignified and simple cork tissues. Cortex appearances broad cortical zone made up of roundish to oval thick walled parenchymas. Stone cells and sclereids found scattered either isolated or in groups. The phloem composed of sieve tubes with compound sieve plates, companion cells, phloem rays and parenchyma. Xylem occupies almost ample central area and consists of vessels, tracheids, fibers, xylem rays and xylem parenchyma. Medullary rays bi to multi seriated. Prismatic and acicular crystals of calcium oxalate and simple and compound starch grains with hilum found throughout the section.

Transverse section of Clerodendrum phlomidis (Linn) F.

(Fig. 2c to 2h) The diagrammatic section of the root shows, outermost cork followed with cortex, phloem and central xylem. Details section shows Cork consisting of 15-20 layers of thin walled tangentially arranged cells having wavy radial conjoining cell walls at places. Cortex demonstrations broad cortical zone made up of roundish to oval thick walled parenchyma embedded with prominent layer of Stone cells and sclereids especially three-sided thickened stone cells and sclereids found scattered either isolated or in groups almost filled with prismatic crystals of calcium oxalate. The phloem composed of sieve tubes with compound sieve plates, companion cells, phloem rays and parenchyma encircling central xylem. Xylem occupies central area and consists of vessels, tracheids, fibers, xylem rays and xylem parenchyma. Medullary rays mostly biseriated. Prismatic and acicular crystals of calcium oxalate and simple and compound starch grains with hilum found throughout the section.

Organoleptic characters and microscopic particulars:

Clerodendrum serratum L. Root: powder was whitish brown in colour, bitter taste, strong aromatic odour and fibrous in touch. (Fig. 3a)

The presence of simple and compound starch grain, prismatic & acicular crystal, bordered pitted vessel, simple & compound starch grain with hilum, stone cell & sclereid and group of fibers observed during the powder microscopy of *Clerodendrum serratum* L. (Fig. 3b to 3j)

Clerodendrum phlomidis L. Root: powder was Creamish yellow in colour, astringent taste, Aromatic odour and fibrous in touch. (Fig. 4a)

Powder microscopy of *Clerodendrum phlomidis* L. found Starch grains, Prismatic crystals, Pitted vessel, Bordered Pitted vessel, fibers, Stone cells and sclereid, brownish coloured cork cell in surface view was observed. (Fig. 4b to 4j)

The study reveals that both the species shows almost similar characters. However, it can be differentiated by some characters. Macroscopically, the colour, size, and shape of the roots are the only the differentiating characters of the both species. Differentiating key characters of both species observed as, in *Clerodendrum serratum* L. bi-multi seriated medullary rays, bordered pitted vessel and simple & compound starch grains with hilum while mostly bi-seriated medullary rays, simple & compound starch grains pitted vessel in *Clerodendrum phlomidis* L. root. The peculiar character like, three-sided thickened stone cells & sclereids was observed only for *Clerodendrum phlomidis* L. root. Starch grains, Prismatic crystals, Bordered Pitted vessel, fibers, Stone cells and sclereid were observed commonly in both the species root powder.(Fig. 3&4)

Phytochemical analysis

All the physicochemical analysis except Loss on drying were comparatively within negligible limits. Loss on drying in raw sample of *Clerodendrum serratum* (Linn) Kutz. Root was found 6.25% w/w. Which is significantly more than that of *Clerodendrum phlomidis* (Linn) F. (3.97% w/w) sample. It is suggested that *Clerodendrum serratum* (Linn) Kutz. Root having more water content than other sample of *Clerodendrum phlomidis* (Linn) F. (Table 2) Preliminary

phytochemical analysis revealed that the presence of metabolic like glycoside, carbohydrates, steroids, flavonoids and tannins in both the samples while alkaloids, protein, amino acids and saponins are absent. (Table 3)

HPTLC profile

The observations are tabulated in table 4. At 254nm; root of Clerodendrum serratum L. found 14 spots where as 8 spots in case of Clerodendrum phlomidis L. respectively. Clerodendrum serratum L. root found 7 spots and also Clerodendrum phlomidis L. found 7 spots at 366 nm. Similar spots of Rf values 0.09 and 0.43 were detected in both the samples (Fig5). Common R_f values like 0.09 and 0.30 at 254nm, 0.09, 0.30 and 0.43 at 366 nm while 0.09 and 0.30 were found for both the samples at both the wavelengths observed similar indicating the presence of analogous type of components in both the species.(Fig. 5) This indicates the presence of same common chemical components as per the observed spots in different wavelengths like visual, 254nm and 366nm respectively, areas and graphical curves. This similarity might be present due to same family or genus of the plant materials. (Table 4)

Table 1: Macroscopic character of C. serratum and C. phlomidis Root (fig. 1a, 1b and 2a, 2b)

Average length	2 cm to 4 cm	8 cm to 10 cm	
Touch	smooth or longitudinally striated or wrinkled, older	root bark thin, easily peal-able, tough, rough due to	
	somewhat rough	exfoliation, longitudinal striated in old one	
Colour	Externally pale reddish brown easily peal-able outer skin, Internally yellowish brown	light brownish ashy in colour, inner surface whitish- yellow	
Odour	Aromatic	Aromatic	
Taste	Bitter, Astringent	Astringent	
Fracture	Short	Hard with sound, Fibrous	

Table 2: Physicochemical parameters of C. serratum and C. phlomidis root powder

Sr. no	Physico -chemical Parameters	C. serratum L	C. phlomidis L
1	Loss on drying(%w/w)	6.25	3.97
2	Ash Value(%w/w)	2.10	2.62
3	Water soluble extract(%w/w)	18.68	18.28
4	Methanol soluble extract (%w/w)	16.6	15.24
5	На	6.2	6.5

Table 3: Result of qualitative tests of methanolic extract of *C. serratum* and *C. phlomidis* root.

Sr. no	Active constituent	Test	C. serratum L	C. phlomidis L
1	Alkaloids	Dragendorff's test	-	-
2	Steroids	Steroids Salkowaski reaction +		+
3	Amino acids	Ninhydrin test	-	-
4	Carbohydrates	Molisch's test	+	+
5	Flavonoids	Lead acetate	+	+
6	Tannins	Lead acetate	+	+
7	Glycosides	Keller Killiani test	+	+
8	Saponins	Foam test	-	-

[&]quot;+": Positive, "-": Negative

Table 4: HPTLC analysis of methanolic extract of *C. serratum* and *C. phlomidis* root.

Sample	Solvent	254 nm (Short UV)		366 nm (Long UV)	
	system	No. of spots	Rf values	No. of spots	Rf values
C. serratum L	Toluene: Ethyl acetate:	14	0.02, 0.09, 0.15, 0.22, 0.31, 0.39, 0.45, 0.54, 0.61, 0.67, 0.72, 0.84, 0.91	7	0.02, 0.09, 0.24, 0.31, 0.36, 0.43, 0.61
C. phlomidis L	Formic acid (7:3:0.3)	8	0.03, 0.10, 0.16, 0.30, 0.52, 0.70, 0.83, 0.90	7	0.2, 0.09, 0.14, 0.30, 0.43, 0.42, 0.84

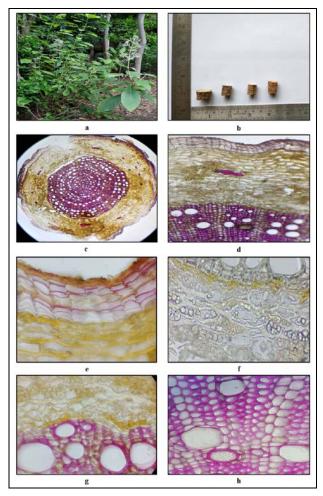
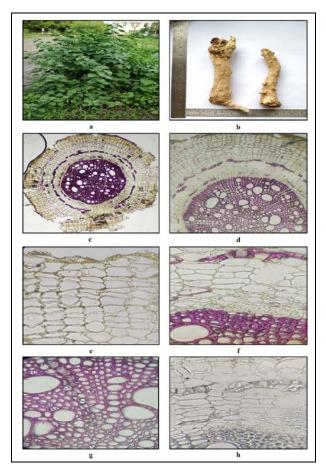
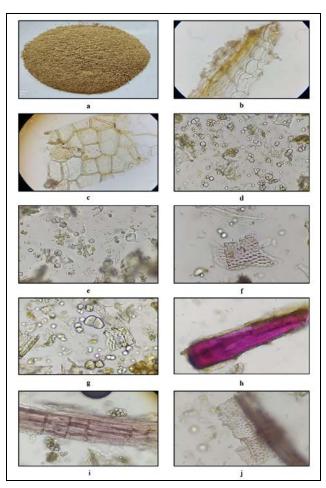


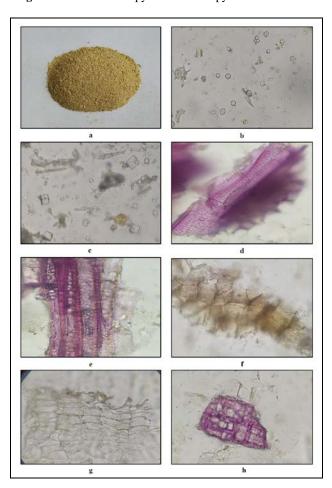
Fig 1: Macroscopic and study of C. serratum L.



 $\textbf{Fig 2:} \ Macroscopic \ and \ Microscopic \ Study \ of \ C. \ Phlomidis \ L.$



 $\textbf{Fig 3:} \ Powder \ Macroscopy \ and \ Microscopy \ of \ C. \ serrratum \ L.$



 $\textbf{Fig 4:} \ \ \textbf{Powder Macroscopy and Microscopy of C. phlomidis L.}$

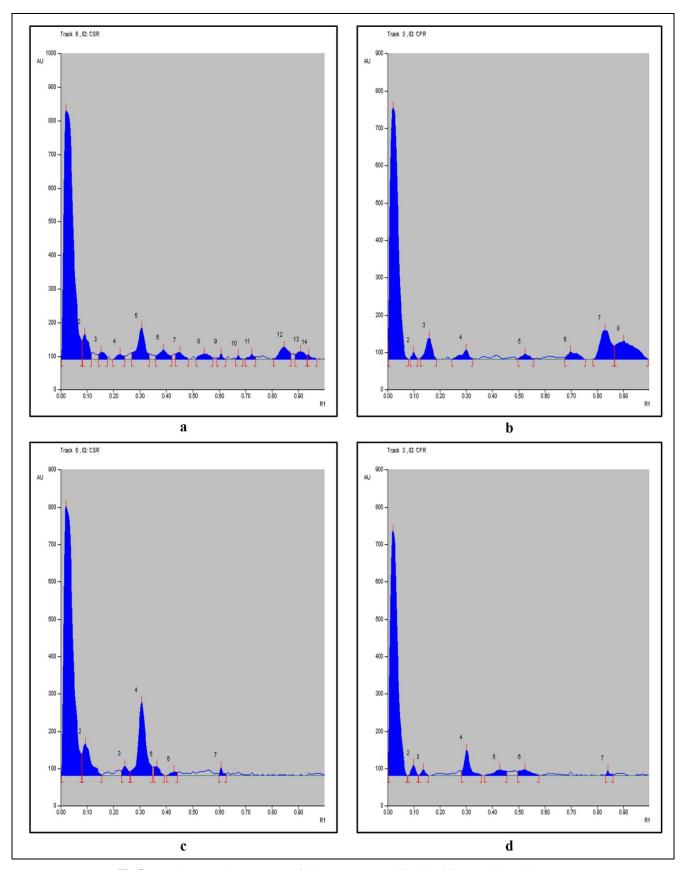


Fig 5: Densitometry chromatogram of C. serratum L. and C. phlomidis L. at 254 and 366 nm.

Conclusion

Bharangi is Clerodendrum serratum (Linn) Kutz. And Tarkari; Clerodendrum phlomidis (Linn) F. according to standard references API and database on medicinal plants belonging to the same genus and family Lamiaceae. According to conducted study of both plants, concluded that

both the species shows the same *Katu*, *Tikta Rasa*. Both the plants have virtually similar morphological and microscopical characters as observed in pharmacognostic study. Consequently, it can be decisively concluded that *Clerodendrum phlomidis* (Linn) F. may be used in the scarcity of *Clerodendrum serratum* (Linn) Kutz. Based on

performed pharmacognostical study and physicophytochemical study.

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