

Pharmacognostic and Phytochemical Characteristics of the Aerial Part of *Salvia moorcroftiana* Wall. ex Benth. Growing Wild in Kashmir Valley, India

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ABSTRACT

Introduction: *Salvia moorcroftiana* Wall. ex Benth. belongs to genus *Salvia*, which is one of the largest genera from Lamiaceae family, which comprises about 900 species and is widely distributed in Kashmir valley. It is used medicinally in guinea worm infection, itching, colic, dysentery and boils. **Objective:** The present study deals with Pharmacognostic parameters of the aerial part of *Salvia moorcroftiana* Wall. ex Benth. **Materials and Methods:** The aerial part of *Salvia moorcroftiana* Wall. ex Benth was collected, shade dried for about 3 weeks and powdered by using mechanical grinder and the powdered aerial part of plant material was evaluated for Pharmacognostic parameters by standard methods. The various fractions of *Salvia moorcroftiana* were subjected to preliminary phytochemical screening for the presence of various Phytoconstituents. The microscopy of the aerial part of *Salvia moorcroftiana* Wall. ex Benth revealed the presence of stomata, covering trichome, calcium-oxalate crystals and multicellular-headed glandular trichome. **Results:** Proximate analysis of the aerial part of *Salvia moorcroftiana* showed that the dried plant powder has 6.5 % total ash value, 1.0 % acid insoluble ash value, 0.2 % sulphated ash value.

Loss on drying was found to be 5.4 %. The fractions of *Salvia moorcroftiana* were found to contain various phytoconstituents. Fluorescence analysis of the plant powder showed the behaviour, when treated with different chemical reagents. **Conclusion:** The current study showed the microscopical characters, the preliminary phytochemical screening and the proximate analysis of the aerial part of *Salvia moorcroftiana*. Information collected from such studies can be used as benchmark in the quality control of this plant as a herbal medicine for treatment of various disease.

Key words: *Salvia moorcroftiana*, Microscopical Characters, Proximate Analysis, Fluorescence Analysis, Phytochemical Screening.

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DOI : 10.5530/phm.2019.1.7

INTRODUCTION

Genus *Salvia* (sage) is the grand member of family *Lamiaceae* comprising about 900 species, across the world.^{1,2} Generally, the plants are 30-15cm of height, herbaceous and perennial rarely biennial or annual with multicolored magnetic flowers. The name *Salvia* comes from Latin word for health (*Salvare* or heal). All over the world, the numerous species of *Salvia* have been used traditionally for the treatment of bronchitis, asthma, cough, digestive and circulation diseases, mouth and throat infection, depression, excessive sweating, skin and other disease.^{3,4} Some species also exhibiting antidiabetic,⁵ antitumor,⁶ antibacterial⁷ and antioxidant properties.⁸ *Salvia moorcroftiana* Wall. ex Benth. generally known as "Kallijari" is herbaceous perennial, occurs in temperate Himalaya from Kashmir to Kumaun. It is of the height of 40-90cm, with stems quadrangular and branched from base. *Salvia moorcroftiana* grows between 5000-9000 feet elevation on disturbed areas and open slopes. *Salvia moorcroftiana* bears large long-stemmed basal leaves with a toothed margin that appear to be covered with white wool. While as flowers of this plant are held in a hairy calyx with showy green veined bracts adding to the plants charm.⁹ In Kashmir valley, the leaves of *Salvia moorcroftiana* Wall. ex Benth. are used medicinally. Leaves are medicine against itching, guinea worm infection. It is applied in the form of poultice to wounds,¹⁰ while as the seeds and roots of are used in colic and dysentery and also applied to boils, used as emetic and against cough and also used for haemorrhoids.^{11,12} Essential oil is also present in *Salvia moorcroftiana*.¹³ In spite of, the medicinal importance of *Salvia moorcroftiana*, there is lack of information available on the Pharmacognostic parameters for identification and standardization of this species in whole as well as in powder form. The current study is designed at the standardization and monograph development and to evaluate the Macroscopical, microscopical

and Pharmacognostic parameters and phytochemical screening of the aerial part of *Salvia moorcroftiana*.

MATERIALS AND METHODS

The aerial part of *Salvia moorcroftiana* Wall. ex Benth. was collected from the Awantipora- Lethpora area, Pulwama Jammu and Kashmir, India. This plant was identified and authenticated by Prof. Akhtar H. Malik, Curator Centre for Biodiversity and Taxonomy (CBT) Department of Botany, University of Kashmir under specimen voucher no. 1173 KASH. A sample specimen of collected material was deposited in herbarium for future references.

Reagents

For this work, the reagents used were all of analytical grade purchased from Central Drug House (P) LTD. Bombay, India.

Macroscopical and Microscopical Evaluation

Macroscopical and Microscopical evaluation of the aerial part of *Salvia moorcroftiana* Wall. ex Benth. were studied according to the methods described in Trease and Evans Pharmacognosy.^{14,15} The Macroscopical and microscopical evaluation of *Salvia moorcroftiana* was taken to prevent adulteration and mistaken in selection procedures of raw medicinal material from genus *Salvia*.

Preparation of Extracts

The fresh air-dried aerial part of *Salvia moorcroftiana* was powdered by using mechanical grinder. The powdered sample of plant material was then subjected to hydroalcoholic extraction by using soxhlet extractor

for 72 hr. The hydroalcoholic extract was concentrated under reduced pressure using rotary vacuum evaporator. After extraction, the dried hydroalcoholic extract was sequentially fractionated with different organic solvents such as Hexane DCM, Ethyl acetate and Butanol in the increasing order of polarity by using separating funnel. The dried fractions were then preserved in air tight glass containers for further use.

Proximate Analysis

Proximate Analysis was carried out on the aerial part of *Salvia moorcroftiana* for the evaluation of various physicochemical parameters such as extractive value (hot and cold),¹⁶ loss on drying,¹⁷ total ash value, acid insoluble ash value, sulphated ash value¹⁸ and PH determination (1% and 10%) solution of drug.¹⁹ Fluorescence analysis study of powdered drug material was carried out by treating with different chemical reagents to detect the colour change under UV at 254nm and 366nm and under visible light.^{20,21} The preliminary phytochemical screening was carried out on the Hexane fraction, DCM fraction, Ethylacetate fraction and Butanol fraction of the aerial part of *Salvia moorcroftiana* to determine the presence of various phytoconstituents.^{22,23}

RESULTS

Macroscopical Evaluation

Colour:	Green
Size:	15- 25cm
Taste:	Slightly bitter
Odour:	Characteristic
Shape:	Ovate- elliptical
Texture:	Smooth
Flowers:	Pale blue in colour
Size of flowers:	2.5 cm long
Stem:	1.5-3 ft tall.

Powder Microscopy of Aerial Part of *Salvia moorcroftiana*

When the powdered aerial part of *Salvia moorcroftiana* was examined under microscope, it showed the presence of stomata. In upper epidermis, simple filament multicellular, cone shaped with thickened tuberos sides covering trichome was found. In the upper epidermis of leaf, calcium oxalate crystals were also found. In powder under examination of microscope, it also shows presence of a multicellular-headed glandular trichomes. The microscopical characters are shown in Figure 1.

Proximate Analysis and Phytochemical Screening

For the evaluation of Pharmacognostic parameters of the aerial part of *Salvia moorcroftiana*, the proximate analysis was used as shown in (Table 1). The phytochemical screening of Hexane fraction, DCM fraction, Ethylacetate fraction and Butanol fraction of *Salvia moorcroftiana* showing presence of various Phytoconstituents are shown in (Table 2). The fluorescence analysis of powdered drug treatment with different reagents was studied under visible light and under UV light as shown in (Table 3).

DISCUSSION

Genus *Salvia* has been widely used as medicinal ingredient, a number of studies revealed that the presence of medicinal substances in the aerial part of genus *Salvia*. This study revealed the characteristics of macroscopy, microscopy and Pharmacognostic parameters from the aerial part of *Salvia moorcroftiana*. This information is expected to be a reference for the selection of raw material for the production of crude extract from aerial part of *Salvia moorcroftiana*. According to World Health Organization guidelines, the macroscopic and microscopic characters are the initial steps in the determination of degree of purity and identification of such materials. The present Macroscopical and microscopical observations of the aerial part of *Salvia moorcroftiana*, thus provides the

Table 1: Physicochemical analysis and extractive value of aerial part of *Salvia moorcroftiana*.

Physicochemical parameters	Results	
Total ash value (%w/w)	6.5	
Acid insoluble ash value (%w/w)	1.0	
Sulphated ash value (%w/w)	0.2	
Loss on drying(%w/w)	5.4	
Swelling index(%w/w)	2	
Foreign matter(%w/w)	0.018	
Foaming index	Less than 100	
PH of 1% solution	5.6	
PH of 10 % solution	5.1	
Extractive values (%w/w)	Cold extractive value	Hot extractive value
Ethanollic	2.8	19.5
Aqueous	15.6	26.8

Table 2: Phytochemical Screening of Hexane fraction, DCM fraction, Ethylacetate fraction and Butanol fraction of *Salvia moorcroftiana*.

TESTS	Inference	Hexane fraction	DCM fraction	Ethylacetate fraction	Butanol fraction
CARBOHYDRATES					
Molish's test	Violet ring	++	++	+++	+++
Fehling's test	Brick red ppt	--	++	++	+++
Barfoed's test	Brick red ppt	++	++	++	++
Selwinoff's test	Pink colour	+	+	+	+
TANNINS					
5%FeCl ₃	Yellow colour	+	+	+	+
Lead acetate	White ppt	-	-	++	++
SAPONINS					
Foam test	Foaming	-	-	-	-

Table 2: Con'

Froth test	Frothing	-	-	-	-
FLAVONOIDS					
Shinoda test	Pink colour	-	-	+++	+++
PHENOLICS					
1%FeCl ₃	Bluish colour	+	-	++	++
ANTHRAQUINONE GLYCOSIDES					
Borntrager's test	Pink Colour	-	-	++	++
CARDAIC GLYCOSIDES					
Keller killiani Test	Brown ring at junction	+	++	++	++
Legal test	Pink colour				
TERPENOIDS					
Salkowski's test	Golden yellow ring at junction	++	++	++	++
PHYTOSTEROLS					
Liebermann's test	Brown ring at junction	++	++	++	++
ALKALOIDS					
Dragendroff's reagent	Orange ppt	-	-	-	-
Mayer's reagent	Cream ppt	-	-	-	-
PROTEINS					
Ninhydrin test	Purple colour	+++	+++	+++	+++
Biuret test	Blue colour	+	+	+	+

Table 3: Fluorescence Analysis of Aerial Part of *Salvia moorcroftiana*.

Treatment	Day Light	UV (254nm)	UV (366nm)
Powder as such	Greenish brown	Faded green	Dark green
Powder treated with distilled water	Light grey	Light grey	Dark grey
Powder treated with GAA	Brownish black	Brownish black	Cream colour
Powder treated with conc. HCl	Yellow green	Brownish black	Brownish black
Powder treated with conc. HCl + H ₂ O	Brownish yellow	Dark yellow	Cream yellow
Powder treated with Pet. ether	Light green	Light brown	Bluish white
Powder treated with 10% NaOH	Faded brown	Brownish black	Brown
Powder treated with methanol	Yellow orange	Brown	Whitish brown
Powder treated with ethylacetate	Yellow green	Dark brown	Dark brown
Powder treated with conc. H ₂ SO ₄	Black	Black	Cream color
Powder treated with conc. H ₂ SO ₄ + H ₂ O	Black brown	Black brown	Dark brown
Powder treated with picric acid	Yellow	Dark yellow	Light yellow
Powder treated with 5% FeCl ₃	Brown orange	Orange	Dark orange
Powder treated with chloroform	Yellowish orange	Dark yellowish orange	Dark orange
Powder treated with HNO ₃	Brown red	Dark red	Light red
Powder treated with HNO ₃ + H ₂ O	Black brown	Black brown	Dark brown
Powder treated with 5% Iodine	Pink red	Dark red	Dark red

useful information for quality control parameters for the crude drugs. The ash value was used to detect the presence of foreign matters in the sample such as sand and soil. The extractive value was used to find out the amount of active constituents. Loss on drying determine the amount of moisture as well as volatile content present in a drug. PH determination reveals the concentration of acidic and basic compounds presents in the extract. The fluorescence is an important phenomenon exhibiting by various chemical constituents present in the plant material and

it is an important Pharmacognostic evaluation parameter. The results of phytochemical screening of various fractions of aerial part of *Salvia moorcroftiana* showed the presence of various Phytoconstituents such as flavonoids, phenols, tannins, terpenoids, phytosterols, carbohydrates etc. the polyphenols and flavonoids are natural antioxidants and possess other pharmacological activities such as anticancer, anti-inflammatory, antianxiety, antimicrobial activities.

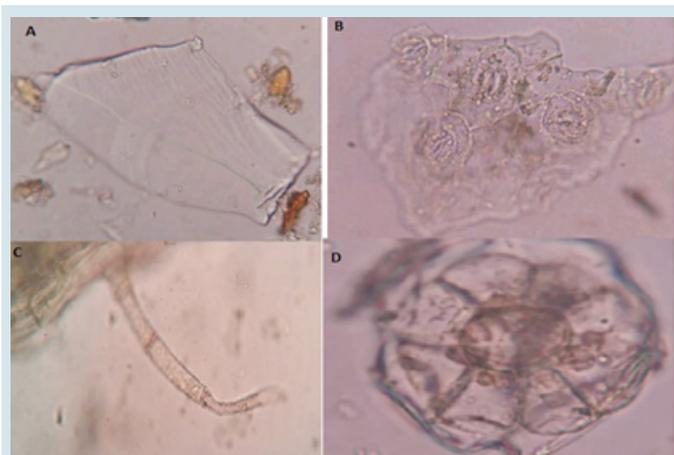


Figure 1: Microscopic Characters of *Salvia moorcroftiana* Observed under Microscope, (a) Calcium oxalate crystals, (b) Upper epidermis in surface view showing stomata, (c) Upper epidermis showing covering trichome and (d) Multicellular-headed glandular trichome in side and surface view.

CONCLUSION

Characteristic macroscopy and microscopy of *Salvia moorcroftiana* (Aerial part) in this study are suggested to be a reference in the selection process of raw materials for the preparation of herbal medicines containing aerial part of *Salvia moorcroftiana*. Pharmacognostic parameters and phytochemical screening were also suggested to be a benchmark for the quality control of the aerial part of *Salvia moorcroftiana*.

ACKNOWLEDGEMENT

The authors acknowledge the support of: Department of Pharmaceutical Sciences (DOPS), University of Kashmir, Srinagar, Jammu and Kashmir, India, for providing the necessary facilities to conduct this study.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

DCM: Dichloromethane; UV: Ultraviolet.

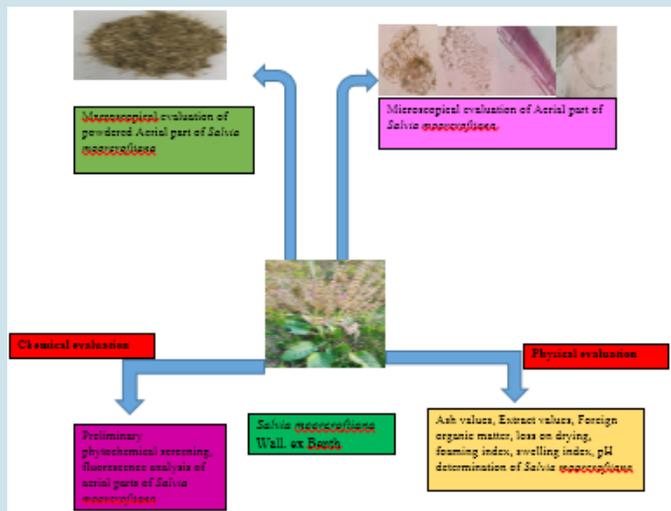
REFERENCES

1. Senatore F, Arnold NA, Piozzi F. Chemical composition of the essential oil of *Salvia multicaulis* Vahl. var. *simplicifolia* Boiss. J Chromatogr A. 2004;1052(1-2):237-40.
2. Senatore F, Arnold NA, Piozzi F, Formisano C. Chemical composition of the es-

sential oil of *Salvia microstegia* Boiss. et *Balansa* growing wild in Lebanon. J Chromatogr A. 2006;1108(2):276-8.

3. Sonboli A, Babakhaniand B, Mehrabian AR. Antimicrobial Activity of Six Constituents of Essential Oil from *Salvia*. Z Naturforsch. 2006;61(3-4):160-4.
4. Yousefzadi M, Sonboli A, Ebrahimi SN, Hashemi SH. Antimicrobial Activity of Essential Oil and Major Constituents of *Salvia chloroleuca*. Z Naturforsch. 2007;63(5-6):337-40.
5. Hitokato H, Morozumi S, Wauke T, Saiki S, Kurata H. Inhibitory effects of species on growth and toxin production of toxigenic fungi. App Envir Microbiol. 1980;39(4):818-22.
6. Ulebelen A, Topcu G, Tan N, Lin LJ, Cordell GA. Microstegiol, a rearranged diterpene from *Salvia microstegia*. Phytochemistry. 1992;31(7):2419-21.
7. Lin LZ, Wang XM, Huang Y, Cordell GA. Antibacterial, antituberculous and anti-phlogistic activities. Phytochemistry. 1989;28:3542-3.
8. Dobrynin VN, Kolosov MN, Chernov BK, Derbentsev NA. Antimicrobial substances of *Salvia officinalis*. Khim Prir Soedin. 1976;1(5):686-7.
9. Betsy C, Barner CD. The New Book of Salvias. Timber Press. 2003;199. ISBN 978-0-88192-560-9.
10. Kirtikar KR. Indian Medicinal Plants (II). Panni, Office Bhuwaneswari Bahadur Ganj, Allahabad. 1918.
11. Bakshi B, Mulchandani NB, Shankar J. A Novel Phenalenone from *Salvia moorcroftiana*. Planta Med. 1986;52(05):408-9.
12. Nadkarani KM. Indian Materia Medica (I). Popular Parakashan Pvt. Ltd., Bombay. 1976.
13. Rather MA, Dar BA, Bhat KA, Shawl AS, Qurishi MA, Dar MY, et al. Monosesquiterpenoid Composition in the Leaves and Flowers of *Salvia moorcroftiana* Wall ex Benth. Growing Wild in Kashmir, India. Journal of Essential Oil Research. 2011;23(4):21-5.
14. Evans WC, Evans D, Trease GE. Trease and Evan's pharmacognosy. 16th ed. Saunders/Elsevier. 2009.
15. Trease GE, Evans WC. Pharmacognosy. International edition. WB. Saunders. 2008;2(3):53844.
16. Chaudhari RD. Herbal drug industry, 1st ed. Eastern Publisher, New Delhi. 1996;498-9.
17. Chase CR, Pratt R. Fluorescence of powdered vegetable drugs with reference to development of a system of identification. Journal of Pharmaceutical Sciences. 1949;38(6):324-31
18. Pharmacopoeia I. Ministry of health and family welfare. Government of India. 1996;2:350.
19. Anonymous. Standardization of Single Drugs of Unani Medicine (Part-II). Central Council for Research in Unani Medicine, New Delhi. 1992.
20. Mukherjee PK. Quality control of herbal drugs: an approach to evaluation of botanicals. New Delhi: Business Horizons Publication. 2002.
21. Kokoski CJ, Kokoski RJ, Slama FJ. Fluorescence of powdered vegetable drugs under ultraviolet radiation. Journal of the American Pharmaceutical Association. 1958;47(10):715-7.
22. Harborne JB. Phytochemical methods In: A guide to modern techniques of plant analysis. 3rd Ed. Chapman and Hall, U.K. ICMR. 1998;56-99.
23. Trease GE, Evans WC. A textbook of Pharmacognosy, London. Bailliere Tindall. 1983;12(193):336.

PICTORIAL ABSTRACT



Zahida Shah, is a Research Scholar in the Department of Pharmaceutical Sciences, University of Kashmir. She is presently doing PhD in Pharmacology and is working on the biological activities of various medicinal plants of Kashmir Himalayas.

SUMMARY

- The current study provides characteristics of aerial part of *Salvia moorcroftiana* as well as phytochemical screening and fluorescence analysis as a benchmark for the quality control of herbal medicines.

ABOUT AUTHORS

Prof. Zulfiqar Ali Bhat, is presently Heading Department of Pharmaceutical Sciences, University of Kashmir. His area of specialization is Pharmacognosy and Phytochemistry. He has more than 20 years of teaching and research experience. His area of research is mainly focused on medicinal plants of Kashmir valley and Ladak region. He has worked on plants having CNS, anti-diabetic, anti-inflammatory, anti-ulcer, anti-hyperlipidemic and hepatoprotective activity. He is also member of many Scientific Journals. Prof. Bhat has number of National and International publications to his credit and has guided a number of PhD scholars.

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