International peer reviewed open access journal

Journal of Medical Pharmaceutical and Allied Sciences

Journal homepage: www.jmpas.com CODEN: JMPACO

Research article

Pharmacognostic and phytochemical assay of *Berberis Aristata* DC roots

Shweta Jain[1], Shalini Tripathi[2], Pushpendra Kumar Tripathi[3]*

[1] Sir Madanlal Institute of Pharmacy, Etawah, Utter Pradesh, India

[2] Rameshwaram Institute of Technology and Management, Dr A P J Abdul Kalam Technical University, Lucknow, Utter Pradesh, India

[3] Institute of Pharmaceutical Sciences, University of Lucknow, Lucknow, Utter Pradesh, India

Corresponding author: Pushpendra Kumar Tripathi ✉ pkt121975@gmail.com, **Orcid Id**: https://orcid.org/0000-0001-9895-2648

Institute of Pharmaceutical Sciences, University of Lucknow, Lucknow, Utter Pradesh, India

Received - 09-01-2023**, Revised** - 10-03-2023, **Accepted** - 12-04-2023 **(DD-MM-YYYY)**

Refer This Article

Shweta Jain, Shalini Tripathi, Pushpendra Kumar Tripathi, 2023. Pharmacognostic and phytochemical assay of *Berberis Aristata* dc roots. Journal of medical pharmaceutical and allied sciences, V 12 - I 3, Pages - 5775 – 5779. Doi: 10.55522/jmpas.V12I3.4800.

ABSTRACT

The study was performed to identify the imperative pharmacognostic details of *berberis aristata* DC roots. The root of the plant was standardized by morphologically and microscopically. Further other parameters chemomicroscopical, physical, phytochemical screening and florescence analysis were performed to determine the diagnostic features of *berberis aristata* DC (*B. aristata*). Macro and microscopic studies were also reported in the present study. Physicochemical parameters including extractive values, ash values, moisture content have been calculated, which showed 0.79 total ash, 0.05 acid insoluble ash, 20 water soluble ash and 0.90 water insoluble ash. The extractive values of *berberis aristata* DC were found to be 3.6, 1.2, 0.6 and 2.3 in ethanol, Pet. ether, acetone, and in water respectively. Phytochemical analysis closely revealed the presence of alkaloids in this plant. These outcomes will further help to standardize, identification and in execution research on this plant.

Keywords - *Berberis aristata,* Ash value, Extractive value, Alkaloids, Glycosides and flavonoids.

INTRODUCTION

The genus *Berberis* occupies a significant place in a variety of traditional medicine systems worldwide for its effective medicinal properties. A.L. Jussieu in 1789, first established Berberides', belongs to the family Berberidaceae, most primitive families of Angiosperms [1]. The genus *Berberis* represents 12 genera with 600 species and around 77 species reported in India [2]. *Berberi's* species have been found to contain a number of alkaloids including berbamine, berberine, jatrorrhizine, palmitine and is tetrandrine. Cortical tissues of roots and stems reported to possess high quantity of alkaloids of *Berberis*. Among old roots, the bark contains large quantity of alkaloids. Furthermore, upper stem parts have low alkaloid concentrations even young leaves are devoid of alkaloids [3]. Berberine, a benzylisoquinoline alkaloid obtained from *Berberis* (Berberidaccae) species. It is isolated from the bark, stem, rhizome, and roots of the plants of the genus *Berberis* [4] including *berberis aristata, berberistinctoria, berberis petiolaris, berberis vulgaris, berberis aquifolium, berberis thunbergii* and *berberis asiatica* [5,6].

Berberis aristata DC. (Berberidaceae) is traditionally used herb reported in all early scriptures of Ayurveda, Charka and Susruta for diverse properties and numerous used in the treatment of diseases [7]. Traditionally this plant is used for its antibacterial, antipyretic, anti-inflammatory, laxative, immunostimulant and antihemorrhagic activity. The plant *berberis aristata* reported to contains aromoline, berberine, berbamine, oxyberberine, palmatine, karachine, oxyacanthine and taxilamine. The root of *berberis aristata* contains aromoline berberine, berbamine, columbamine, dehydrocaroline, epiberberine, jatrorhizine, karachine dihyrokarachine, oxyberberine, oxycanthine, palmatine and taximaline. Additionally, 1-O methyl pakistanine, pakistanine, pseudopalmatine chloride and pseudoberberine chloride are alkaloids, isolated from *Berberis*

aristata. A bisbenzlisoquinoline or simple is quinoline alkaloid was isolated from *Berberis aristata* [7]. Berberine is the major alkaloid present in *berberis aristata* with yield of 2.23% followed by palamatine [8]. Aqueous extracts of this plant topically applied and resulted in significant reduction in inflammation. So, this plant is commonly used in traditional medicine in various inflammatory disorders. Additionally, plant is used in ophthalmic complication, malaria, skin diseases and diarrhoea. Pharmacological activities of this plant including antimicrobial and anti-inflammatory make it apposite herb in the treatment of variety of disease [9, 10]. Hence the present research work was performed to assess numerous pharmacognostic parameters including microscopy, macroscopy, phytochemical and physicochemical studies of this valuable plant *berberis aristata* DC.

MATERIALS AND METHODS

Plant material

Fresh roots of the *berberis aristata* DC. was collected from the local area of village Malera, District Almora (Uttarakhand) India. Identification and confirmation were done by Dr. P.K. Khare, Department of Botany, Dr. H.S. Gour Central University, Sagar (M.P.) (herbarium no. Bot./H/112/196119) India.

Chemicals and Reagents

All the reagents including Dragendroff's reagent, Mayer's reagent, Hager's reagent, Wagner's reagent and chemicals i.e., ethanol, methanol, sodium hydroxide, chloroform, glycerine, and phloroglucenol were analytical grade and purchased from commercial chemical supplier (Sigma-Aldrich,, Mumbai, India).

Macroscopic characters of root of *B. aristata* DC.

The macroscopic characters like size and shape, colour, surfaces, texture, odour and taste for the fresh root were noted.

Transverse section of root of *B. aristata* DC.

Microscopic evaluation was performed using transverse sections of fresh root which was treated with chloral hydrate, mounted with glycerine and viewed under the compound microscope. The various types of tissues like epidermal tissues, ground and vascular tissues such as epidermal cells (upper and lower region), covering trichomes, xylem, phloem, stone cells were observed [11].

Chemo microscopic examination

Examination of the powdered drug was carried out for the determination of lignified cells, starch grains, stone cells, xylem and phloem calcium oxalate crystals and fibers by using method reported by khandelwal and sethi (2013) [12] . In brief plant roots were dried at 60ºC (4-6 hrs) and grounded using electric grinder and prepared powder was passed over 60 no sieve. Treated with different staining reagents like phloroglucinol, hydrochloric acid solution, glycerin and iodine solution. The prepared slides were examined for the determination of starch grains, xylem and phloem vessels, cork cells, stone cells, fibers and other cell contents present in the drug [13].

Fluorescence analysis

Powdered roots were analysed under day/visible light and ultraviolet light (Toshiba India) by treating the drug with different reagents or chemicals for successful fluorescence analysis [14]. The powder was treated with acetone, alcohol, benzene, chloroform, ethyl acetate, hexane, methanol, N HCl, 50% H_2SO_4 and aqueous or alcoholic 1N NaOH.

Physicochemical parameters

The total ash was determined using previously ignited and weighed silica crucible, and adds 5 gm of the powdered drugs. It was then burned in a muffle furnace at a temperature (7000 ºC) until it was carbon-free, after which it was cooled and weighed. The material that was air dried was used to calculate the total ash percentage [15].

The obtained total ash was heated in 10 ml of distilled water for 5 minutes. After filtration, the insoluble material was collected on ash less filter paper in silica crucible. Further it was washed with hot water and then ignited at moderate temperature to maintain a constant weight. The weight of the ash was deducted from the weight of the weight of the insoluble material. With reference to the amount of ash consumed, the percentage of water-soluble ash was calculated with 25 ml of 10% HCl, the total amount of ash was heated for five minutes. Insoluble ash was gathered either on an ash-free filter paper or in a silica crucible. It was fired, weighed, and then washed with hot water. The weight of the ash was deducted from the weight of the insoluble material. The acid-insoluble ash is depicted by the weight difference. With reference to the amount of ash collected, the percentage of acid insoluble ash was computed.

Using a maceration procedure, water, ethanol, acetone, petroleum ether was used to extract the dry powdered plant material of *berberis aristata* DC. A dry 250 ml conical flask was filled with 2 g of the plant material that had been roughly powdered and weighed in a weighing bottle. The flask was then successively filled with 30 ml of each of the various solvents. The flasks were corked and left at room temperature for 24 hours while being periodically shaken. The mixtures were transferred to a 50 ml measuring cylinder after being filtered through Whatmann No. 1 filter paper. The filtrate was put into weighed Petry plates after it had been obtained. By allowing the filtrate to sit till the solvent completely evaporated, the resulting extracts were concentrated until dry. The resulting values were calculated by using given formula·

$$\text{Extractive value (\%)} = \frac{\text{Weight of dried extract (obtained)}}{\text{Weight of plant material (actual) X 100}}$$

Hot air oven was used to measure the moisture content of powdered drug. Accurately weighted 10 g of drug was placed in oven at the range of temperature (100 to 110 ºC) and collected in a

desiccator till weight get constant, and weighed after temperature was adjusted to a continuous range of weight loss was used as a gauge for moisture content.

Preliminary phytochemical screening

An effectual phytochemical screening of the plant material is needed for recognizing plant active constituents. Phytochemical extracts were prepared using successive solvent extraction method. In brief powdered drug (60-80 mesh size) was extracted with different solvents like water, ethanol, chloroform, petroleum ether and ethyl acetate. The prepared extracts were screened using the standard procedure for identifying the presence of various chemical constituents, including alkaloids (Mayer's reagent and dragendorff's reagent), flavonoids (Shinoda test), glycosides (Fehling solution test), carbohydrates (Molisch's test), tannins (tannic acid and ferric chloride test) and terpenoids (Salkowski test) [16,17].

RESULTS AND DISCUSSION

Macroscopic evaluation of root

Macroscopically, external surface of the root was corky, grayish brown in color, up to 6 cm or more in diameter (Figure 1). Internally roots are thick, woody yellowish brown, enclosed by a thin brittle bark. Bark is inside pale brown, coarse, closely and rather deeply furrowed. Roots are cylindrical in shape with more or less knotty and cut surface bright yellow, coarse, fibrous with small fine ridges; growth rings discernible, fracture very hard, texture short, odorless and bitter in taste (Table 1).

Figure 1: Root of *B. aristata* DC

Table 1: Morphology of root of *B.aristata* DC.

Macroscopic parameters	Observations
Surface	Corky
Size	Up to 6 cm or more
Shape	Cylindrical
Colour	Inner surface bright yellow Outer surface pale brown
Taste	Bitter
Odor	Odorless
Fracture	Irregular
Texture	Short

Microscopic evaluation of the root

The microscopic examinations help in the diagnosis as well as differentiation and identification of specific species of herbs or plants. The microscopic examination of fresh root was performed and results revealed that the root is circular in outline. Cortical zone contains yellow-colored berberine alkaloids. Calcium-oxalate crystals and starch of are present in medullary rays. The solitary vessels in groups of 2 or 3 were present. The narrow central zone occupied by tracheid and few vessels. A radially arranged cortical cells with vary in size and shape were observed; while stone cells are in groups (Figure 2).

Chemo microscopic examination

The dried root powder is yellowish brown in color with bitter in taste, and lacking of specific odor. Microscopic examination of powdered drug revealed that plant root contains a number of cell contents such as medullary parenchyma cells, stone cells, fragments of rectangular cork cells, pitted, and reticulate vessels, cortical parenchymatous cells, pericyclic fibers, and spiral tracheids with bordered pits, starch and other cell structures (Figure 3, 4, 5 and 6).

Figure 2: Transverse section of fresh root of *B. aristata* DC

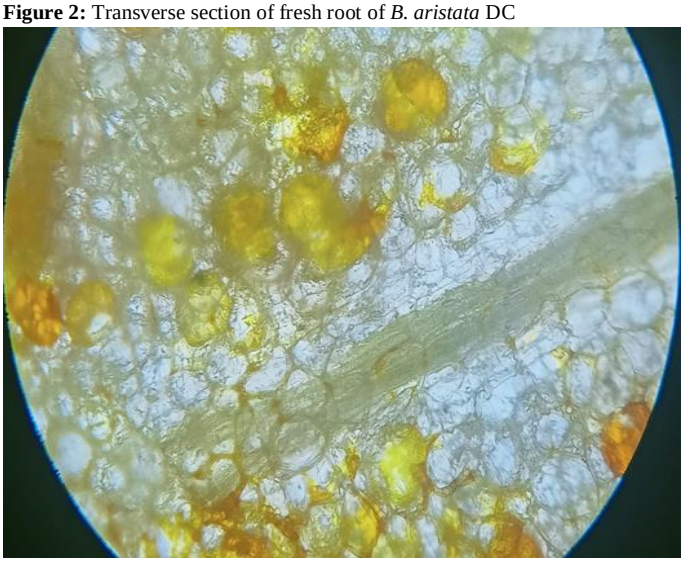

Figure 3: Medullary parenchyma cells

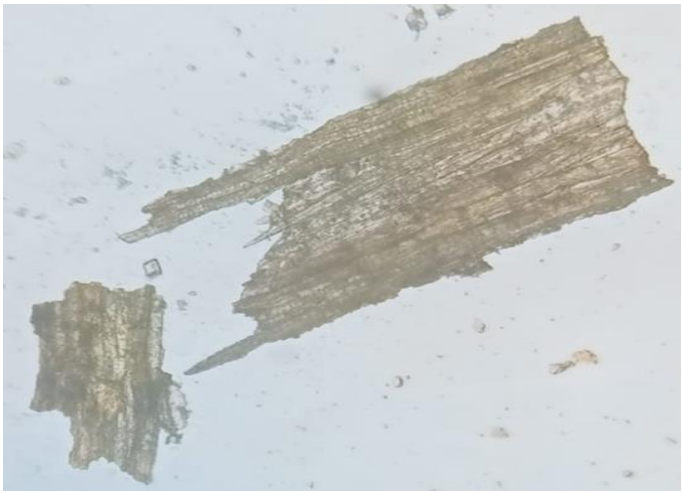

Figure 4: Group of stone cells

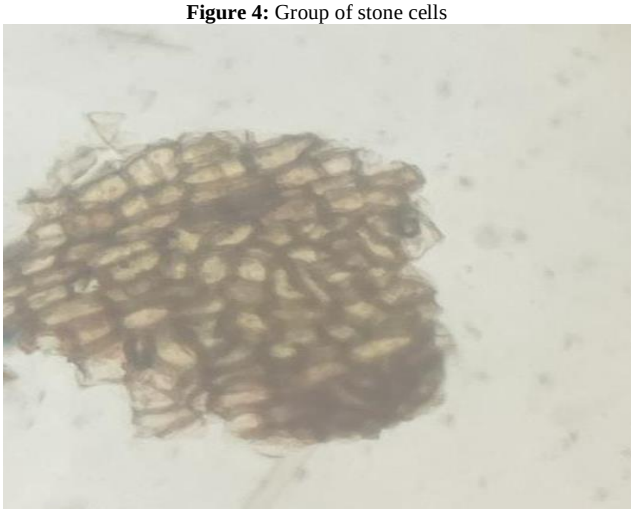

Figure 5: Cork cells

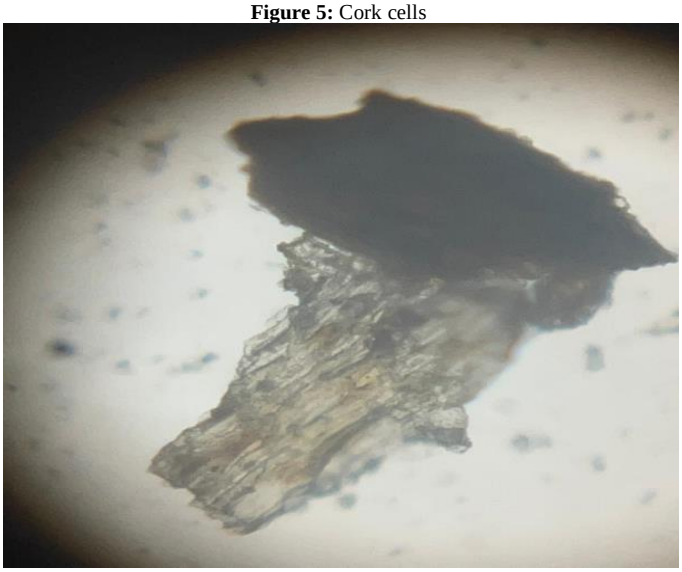

Figure 6: Phloem fibers

Physicochemical parameters

Various physicochemical parameters of powdered drug were investigated (Table 2). Moisture content of drugs may prone to microbial growth however it might be present at lowest level to reduce the growth of fungi, yeast and bacteria on storage. The ash value of dried root powder is reported in Table 2.

It indicates the presence of oxalate, carbonate and silicate impurities. The acid insoluble ash contains silica as impurity of earthy material. The number of inorganic elements can be estimated by water soluble ash determination.

Table 2: Physicochemical parameter of powdered drug

Physiochemical parameter Mean (%w/w)		
Moisture content	Loss on drying at 110˚ C	4.37
Ash values	Total ash	0.79
	Acid insoluble ash	0.05
	Water soluble ash	20
	Water insoluble ash	0.90

Determination of extractive values by cold maceration method

Extractive values give gives rough approximation of chemical constituents present. The outcomes of the study are reported in (Table 3)

Table 3: Extractive values of *berberis aristata* DC

Solvents	Extractive value(%w/w)
Alcohol	3.6
Pet. Ether	1.2
Acetone	0.6
Water	2.3

Florescence analysis of powdered drug

The results of fluorescence analysis have been reported in Table 4.

Table 4 : Fluorescence analysis of powdered drug and extracts of root.

Reagents	UV at long 364nm.	under ordinary light
As such	brownish yellow	greenish yellow
Powder + Con. HNO_3	black	brown
Powder + Con. HCl	faint green	dark brown
Powder + 5% Iodine solution	light brown	light yellow
Powder + 1N. NaOH in methanol	reddish brown	orange
Powder + 5% $FeCl_3$ Green	light red	light yellow
Powder + 1N. NaOH in H_2O	green	dark brown
Powder + Glacial acetic acid	greenish yellow	blackish brown
Powder + Con. H_2SO_4	dark brown	black
Powder + Picric acid	reddish brown	yellow
Extracts		
Pet. ether	brown	dark brown
Ethanol	green	light green
Acetone	brown	green
Water	bark brown	light brown

Color, consistency, odour, taste and extractive values are shown in Table 5.

Phytochemical analysis

The primary and secondary metabolites, viz. alkaloids, carbohydrates, flavonoids, glycosides, fixed oils, tannins and terpenoids were assayed using standard screening test (Table 6)

Table 6: Preliminary qualitative phytochemical analysis of various extracts of *B. aristata* root

Plant constituents	Alcohol	Pet. Ether	Acetone	Water
Alkaloids	+++	+	+	-
Glycosides	++	-	-	+
Terpenoids	-	+	+	-
Tannins	-	_	_	-
Resins	++	++	+	+
Carbohydrates	+	+	_	+
Flavonoids	++	-	++	_

Table 5: Color, consistency, odour, taste and extractive values of successive solvent extraction of powdered drug

Extract	Colour	Consistency	Odour	Taste	Extractive value (%w/w)
Pet. ether	dark green	S-S	Ch	Bt	1.7
Chloroform	green	S-S	Ch	Bt	0.9
Ethyl-acetate	dark green	S-S	Ch	Bt	2.0
Ethanolic	dark brown	S-S	Pu	Sw	5.9
Direct ethanolic	brown	S-S	Pu	Sw	10.1
Aqueous	brown	S-S	Ch	Bt	0.3

Semi-Solid: S-S; Characteristic: Ch; Pungent: Pu; Bitter: Bt; Sweet: Sw

CONCLUSION

In the present research work a number of prescribed standardization parameters were conducted on *B. aristata* DC roots. Pharmacognostic studies results reveled that plant contains various identifiable important cell structures like fibers, cork cells, sclerieids fibers, xylem, phloem, stone cells etc. Macroscopic, microscopic and chemo-microscopic examination of the *B. aristata* DC roots were done. Phytochemical screening was done to confirm the presence of secondary plant metabolites like alkaloids, glycosides, tannins etc. results reveled that plat contain alkaloids in more amount. These results may help in standardization, identification and in carrying out further research on this plant.

ACKNOWLEDGMENT

Author thanks to Dr. P.K. Khare, Department of Botany, Dr. H.S. Gour Central University, Sagar (Madhya pradesh) for identification and confirmation of plant.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships.

REFERENCES

1. Srivastava S, Srivastava M, Misra A, et al, 2015. A review on biological and chemical diversity in berberis (berberidaceae). Experimental and Clinical Sciences Journal 14, pages - 247-267.
2. Chander V, Aswal JS, Dobhal R, et al, 2017. A review on pharmacological potential of Berberine; an active component of Himalayan *Berberis aristata*. Journal of Phytopharmacology 6(1), pages- 53–58.
3. Greathouse and Rigler, 1940. Isolation of the alkaloids, berberine and berbamine from mahonia swaseyi. Plant Physiology 15(3), pages- 563-564.
4. Koppen LM, Whitaker A, Rosene A, et al, 2017. Efficacy of Berberine Alone and in Combination for the Treatment of Hyperlipidemia: A Systematic Review. Journal of Evidence-Based Complementary & Alternative Medicine 22(4), pages- 956-968.
5. Shigwanb H, Saklania A, Hamrapurkarb PD, et al, 2013. HPLC Method Development and Validation for Quantification of Berberine from Berberis aristata and Berberis tinctoria. International Journal of Applied Science and Engineering 11(2), pages- 203-211.
6. Singh A, Duggal S, Kaur N, et al, 2010. Berberine: Alkaloid with wide spectrum of pharmacological activities. Journal of Natural Products 3, pages- 64-75.
7. Yadav R and Mahalwal VS, 2018. The pharmacognostical studies of Berberis aristata DC. International Journal of Green Pharmacy 12(2), pages- 148-152.
8. Kirtikar KR, Basu BD, 1984.Indian Medicinal Plants. Allahabad: Publshed by Basu LM. 2 (2), pages- 1361-1363.
9. Kumar R, Gupta YK, Singh S, 2016. Anti-inflammatory and anti-granuloma activity of Berberis aristata DC. in experimental models of inflammation. Indian Journal of Pharmacology 48(2), pages- 155-161.
10. Saravanakumar T, Venkatasubramanian P, Vasanthi NS, et al, 2014. Antimicrobial potential of Daruharidra (*Berberis aristata* DC) against the pathogens causing eye infection. International Journal of Green Pharmacy, pages- 153-157.
11. Evans WC, Trease GE, 1985. Pharmacognosy. London: WBS aunders. 14, pages-32-33.
12. Khandelwal KR, Sethi V. 2013. Practical Pharmacognosy Techniques and Experiments. Nirali Prakashan, pages- 1-14.
13. Khandelwal KR, Pawar AP, Kokate CK, et al, 2001. Practical Pharmacognosy.Pune: Nirali Prakashan, pages- 19-153.
14. Khim. 1982. Evaluation of some herbal drugs Systematic approach Fitotherapia. 59(6), pages - 494- 495.
15. Anonymous,1999. The Ayurvedic Pharmacopoeia of India. Gov ernment of India, Ministry of Health and Family Welfare. New Delhi. Part I, Vol III, pages- 235-238.
16. Sailor GU, Ghanshyam P, Ashvin VD, et al, 2010. Pharmacognostical and Preliminary Phytochemical Investigation of Leucas cephalotes (Roth) Spreng. International Journal of Pharmaceutical Research 2, pages -14-21.
17. Jain S, Vaidya A, Khatri P, 2011. Pharmacognostic and phytochemical investigations of the leaves of *Zizyphus Xylopyrus* (retz) willd. International Journal of Pharmacy and Pharmaceutical Sciences 3, pages - 122-125.