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# Pharmacognostic Evaluation of the Root of *Berberis aristata* DC.

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**Abstract** – *Berberis aristata* (family Berberidaceae), known as 'Daruharidra' in Ayurvedic system of medicine, is an important medicinal plant used extensively for treating a variety of ailments in various systems of indigenous medicine. Being an important medicinal plant it is being adulterated and in the absence of any pharmacognostic information it is very difficult to check the adulteration. The present study was therefore, carried out to provide the requisite pharmacognostic details. Morphological, anatomical and phytochemical aspects of *B. aristata* were carried out. Diagnostic features of *B. aristata* root were identified and characterized from the above investigations and presented in the present communication. Some of the diagnostic features of the root drug noted from the anatomical study are patches of pericyclic fibre, pitted sclerieds, berberine containing cells and heterocyclic medullary rays. HPTLC analysis showed three distinct bands of which berberine was identified as the major constituents. The $R_f$ value of other bands was also calculated.

**Key words** – *Berberis aristata*, Daruharidra, Pharmacognosy, Root, HPTLC, Berberine.

## Introduction

*Berberis aristata* DC. belonging to the family Berberidaceae, is one of the most important medicinal plants and used extensively in almost all indigenous systems of medicine known as 'Daruharidra' in Ayurveda, where it is used in the treatment of jaundice, enlargement of spleen (Anonymous, 1948). The root extract commonly known as 'Rasot' is an alternative, deobustruent and are used in skin diseases, menorrhagia, diarrhoea, jaundice and above all in affection of the eyes (Kirtikar and Basu, 1933).

A decoction made from this drug is used as mouth-wash for treating swollen gums and toothache (Anonymous, 1948). Extracts of the fresh root is extensively used as a purgative for children. 50% aqueous alcoholic extract of root is hypoglycemic and anticancerous, used as contraceptive with the extracts of *Iris germanica* & *Terminalia chebula* (Sharma *et al.*, 1993; Sohni *et al.*, 1995). Tripathi (1996) worked out on hepatopathy in goats, and antiamoebic effect of a crude drug formulation with *B. aristata* which is used against *Entamoeba histolytica* and also to treat allergic disorders. A poly-herbal formulation with *B. aristata* and other 5 herbs was evaluated in experimental amoebic liver abscess in golden hamsters and in immunomodulation studies (Sohni *et al.* 1996).

Decoction of root bark is used for treating malarial fevers and claimed to have tonic, antiperiodic and diaphoretic properties (Kirtikar and Basu, 1933). It has been found to be of great value in intermittent and remittent fevers and in debility, diarrhoea and dyspepsia (Bently, 1983). Paste of Root-bark is applied for healing ulcers and a combination with opium, rock salt and alum is considered to be a useful anti-inflammatory agent (Chaterjee, 1994).

Ethnomedical investigation carried out by Chauhan (1978-79) reported that the decoction of root is used in piles, gastric disorders and other allied complaints by Tibetans, and Shah (1971) reported that the tribals of Kumaun region use the same decoction for treating eye troubles and boils.

Gilani (1992) reported that the leaves of *B. aristata* can prevent acetaminophen-induced liver damage. Besides its active constituent berberine, two other protoberberine type alkaloids karachine and taxilamine were isolated from its root (Rastogi and Mehrotra, 1980-84). No detailed work on the pharmacognosy is on record, hence, the present study has been undertaken.

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## Material and Methods

*Berberis aristata* was collected from the Ranikhet (U.P.) region in the month of May from where this drug is traditionally collected. The roots were preserved in 70% Alcohol for histological studies. Hand sections were cut and stained with safranin. Physico-chemical and phytochemical studies like, total ash, acid insoluble ash, tannin and total alkaloi d were made from the shade dried powdered material.

## Results and Discussion

### Observations

**Brief Taxonomic Description of the Plant –** *Berberis aristata* DC. is a large deciduous shrub usually 1.8-3.6 m high. Twigs whitish or pale yellowish brown erect cylindrical, smooth and strongly striate. Blaze 5-7.5 mm, bright yellow with coarse reticulate fibres. Leaves 3.8-10×1.5-3.3 cm, obovate or elliptic, entire or spinous-toothed, base gradually narrowed, with prominent reticulate nerves, glossy dark green above, glossy pale green but not glaucous beneath. Flowers numerous, stalked. Inflorescence a simple drooping raceme, bracts small, linear, acuminate. Sepals 8 or 9, imbricate, oval, petaloid, yellow. Petals 6, in two whorls, strongly imbricate, concave, bright yellow veined with two oval linear glands at the base of the lateral veins. Stamens 6 equal, hypogynous, opposite and slightly shorter than the petals. Ovary simple, 1-celled, with a few erect ovules. Style short, Stigma peltate. Fruit a small berry about 7-10 mm, ovoid or oblong ovoid, blue black with a whitish bloom tipped along with the persistent style and stigma.

**Macroscopic characters of the Root –** The roots are thick, woody, yellowish brown, cylindrical, more or less knotty and covered with a thin brittle bark. Bark is internally pale brown, rough, closely and rather deeply furrowed. Cut surface bright yellow, rough, fibrous with small fine ridges; growth rings discernible, wood diffused porous, fracture hard, texture short; odourless and bitter in taste.

**Microscopic characters of the Root (Plate 1) –** T.S. of mature root is almost circular in outline (Fig. 1). The outer most cork is composed of rectangular 13-15 layer cork cells, followed by 2-3 layered cork cambium. The cortical region is made up of 30-35 layered rectangular parenchymatous cells, which are filled with tannins. Pericyclic fibres in patches,

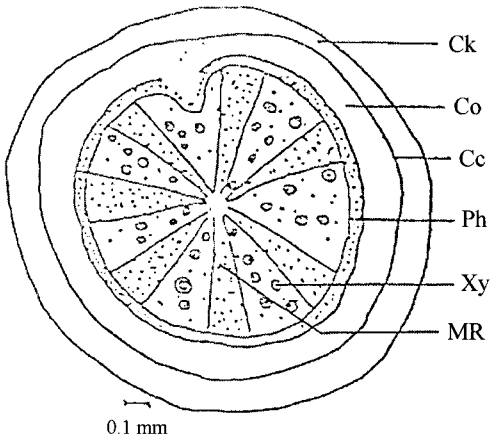


**Fig. 1.** Diagrammatic representation of T.S. of Root.

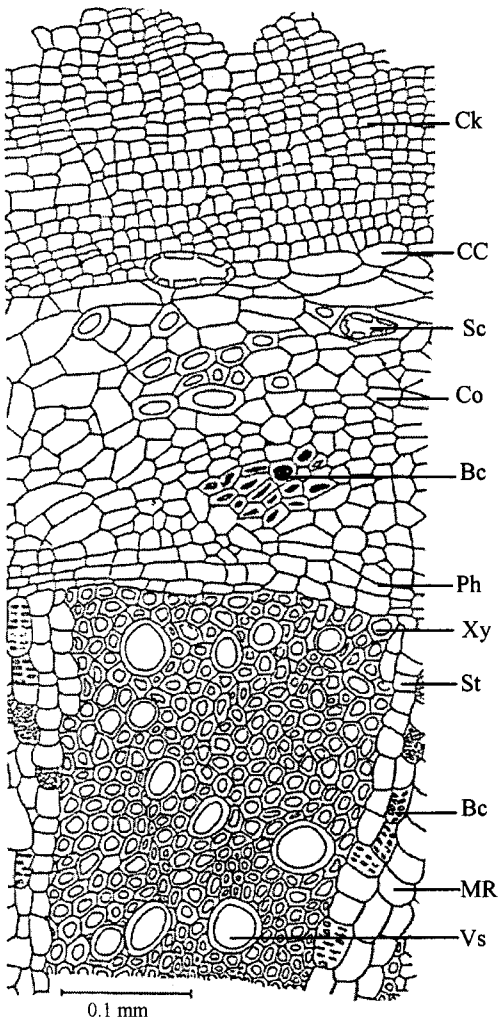


**Fig. 2.** T.S. Cellular of the root.

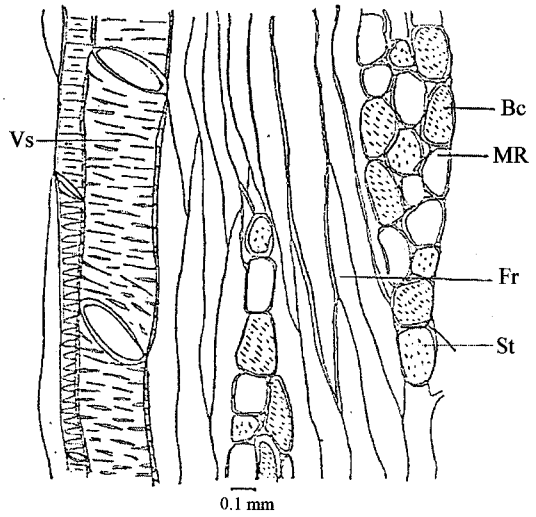


**Fig. 3.** TLS of the Root.

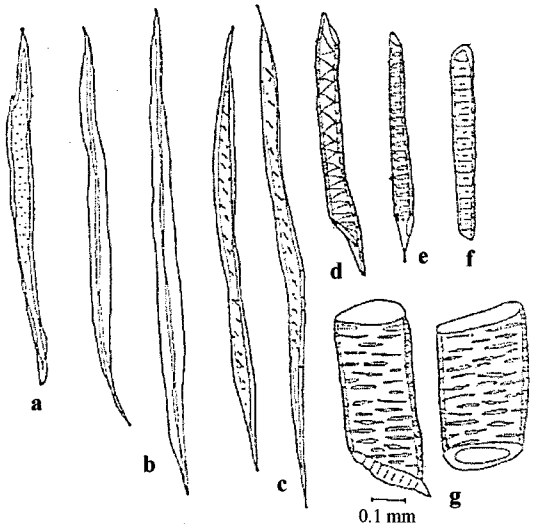


**Fig. 4.** Macerated Elements. a-b. Fibre, c. Tracheidal fibres, d. Vessels with spiral thickenings, e. Vessels with annular thickenings, f. Vessels with scalariform thickenings, g. Tailed vessels with spiral thickenings.

sclerieds mostly solitary or in a group of 2-5 are also observed in cortical region. Some cortical cells are filled with yellow coloured alkaloidal content. Just below the cortex 3-4 celled thick secondary phloem is observed which consists of sieve tubes, companion cells and phloem fibres. Cambium is 2-3 layered. The secondary xylem is 8-10 celled wide and consists of vessels, fibres, tracheids and parenchyma. Vessels mostly solitary or in a groups of 2-3 surrounded by

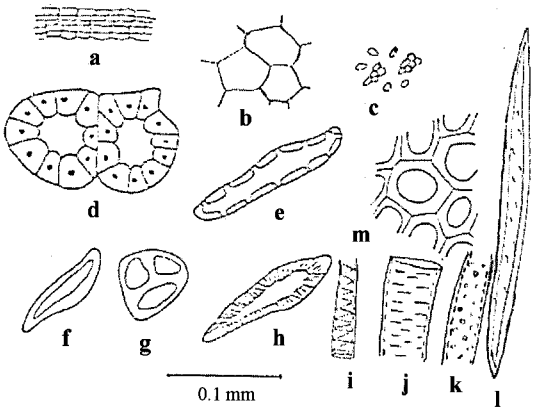


**Fig. 5.** Microscopic details of Powder. a. Cork cells, b. Parenchymatous cells, c. Starch grains, d-h. Scleieds, i-k. Vessels, l. tracheids, m. Pericyclic fibre patches.

parenchyma which is vasicentric. Crushed vessels and fibres are also observed in the center (Fig. 2). The medullary rays are thin, radiating, parenchymatous, heterogeneous and filled with starch and alkaloidal content (Fig. 3). The medullary cells with alkaloidal content are pitted.

On maceration, the vessels (259.72×25.59 μm) with annular, reticulate, spiral, scalariform and some with simple pits are observed and tailed vessels are also observed. The tracheids with bordered pits measuring 491.02×13.06 μm and tracheidal fibres 694.06×12.98 and simple fibres 645.48×14.66 μm are clearly discernible (Fig. 4).

**Study of Powder (Fig. 5)** – The powder of the whole drug is yellowish brown, bitter in taste and without specific odour. The powder was sieved through 40 mesh, cleared in chloral hydrate and mounted in glycerin. On microscopic examination the powder revealed the presence of fragments of rectangular cork cells, cortical parenchymatous cells, pericyclic fibres, stone cells, fragments of spiral, pitted and reticulate vessels, fibres, tracheids with bordered pits and starch in single or compound grains.

The behaviour of the powdered drug with different chemical reagents was also studied as per methods described by Chase and Pratt (1949) and Kokoski *et al.* (1958). Powder when treated with 50% $H_2SO_4$ became fluorescent yellowish light brown in colour, while with acetic acid and iodine water fluorescent yellow in daylight and emits fluorescent yellowish green colour in UV-254, respectively.

**Phytochemical Studies**

Air-dried material was used for quantitative determination of different physico-chemical values and other phytochemical work. The recommended procedures were followed for calculating total ash (Anonymous, 1966), tannin by AOAC spectrophotometric method (Anonymous, 1965) and alkaloid (Siwon *et al.*, 1980). The values obtained are recorded in Table 1.

The known quantity of dried powder was extracted in Soxhlet with hexane, chloroform, acetone, alcohol and water successively (Table 2) and tested for different constituents (Peach and Tracy, 1955) *viz.* steroids and triterpenoids (LB test), flavonoids (Shinodas test), alkaloids (Mayers reagent), tannins (Ferric chloride test) and sugar (Fehling solution test). The study revealed that the triterpenoids are present in hexane and chloroform soluble parts, tannin is only in water soluble part while the resin present in acetone and alkaloids in chloroform, acetone, alcohol and water soluble parts.

A densitometric HPTLC analysis was also performed for the development of characteristic fingerprint profile, which may be used as markers for quality evaluation and standardization of the drug. For this 1 gm powdered root was refluxed for 5 minutes on water bath with 5 ml methanol, filtered and filtrate taken as test solution along with reference berberine (7 µl of each) and was applied on HPTLC precoated silica gel G60 $F_{254}$ Merck glass plates of 20×10 cm with the help of Camag Linomat-IV applicator and eluted the plate to a distance of 6.20 cm at room temperature (19°C) in solvent system *n*-propanol : water : formic acid (90 : 80 : 0.4). The bands in the sample are obtained at $R_f$s 0.19, 0.30 and 0.43, which can be used as identifying markers.

The berberine was identified at $R_f$ 0.30 and its percentage was also calculated (Fig. 6).

**Table 1.** Quantitative Physico-chemical Analysis of Root of *B. aristata*

| S. No. | Parameters | Mean* ( in percent) | S.D. |
|---|---|---|---|
| 1 | Moisture | 42.55 | ±1.2503 |
| 2 | Total ash | 2.18 | ±0.0288 |
| 3 | Acid insoluble ash | 0.05 | ±0.0000 |
| 4 | Alcohol soluble extractive | 9.50 | ±0.0000 |
| 5 | Water soluble extractive | 14.00 | ±0.0000 |
| 6 | Tannin | 0.616 | ±0.0000 |
| 7 | Alkaloid extract (Berberine) | 2.8 | ±0.0000 |

**Table 2.** Successive Soxhlet extractive values

| S. No. | Extractives | Mean* | S.D. |
|---|---|---|---|
| 1 | Hexane extractive | 0.53 | ± 0.0757 |
| 2 | Chloroform extractive | 1.77 | ±0.5071 |
| 3 | Acetone extractive | 6.51 | ±2.0829 |
| 4 | Alcohol extractive | 7.83 | ±1.6911 |
| 5 | Water extractive | 5.96 | ±0.7748 |

* Average of three readings.

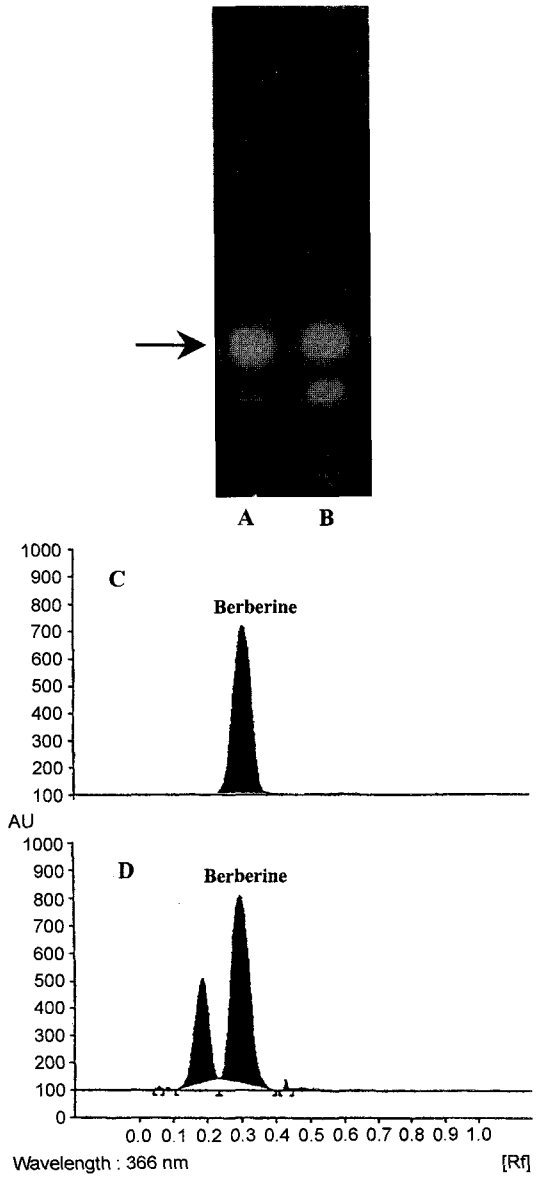


**Fig. 6.** HPTLC Profile of *Berberis aristata* and reference sample (under UV 366). A. Reference sample of Berberine, B. HPTLC profile of *Berberis aristata* Root, C. Chromatogram of reference sample, D. Chromatogram of *B. aristata* Root.

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