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Pharmacognostical and phytochemical investigation of *Barleria prionitis Linn* leaves.

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ABSTRACT

In the present study leaves of *Barleria prionitis Linn* was subjected to pharmacognostical studies such as macroscopic, microscopic and micromeretics parameters were also observed. Physicochemical studies such as ash values, extractive values of plant part were carried out to confirm the identity of plant. Ash values such as total ash, acid insoluble ash and water soluble ash were determined and recorded. Extractive values such as alcohol soluble extractives and water soluble extractive values were also determined. The leaves of *Barleria prionitis Linn* shows the presence of phytoconstituents such as alkaloids, glycosides and tannins.

Key words: *Barleria prionitis*, Microscopy, Pharmacognosy, Photochemistry

INTRODUCTION

Barleria Prionitis L. (Acanthaceae), commonly known, Kuranaka, Korana, Keranaka. A much-branched, prickly shrub, up to 0.6 to 3.0 m in height, found growing throughout the hotter parts of India. It is also commonly grown as a hedge plant in gardens as in Ayurveda, used as kustha it is a febrifuge formulation.

The leaves and stems showed the presence of five iridoid glucosides; three of them, acetyl barlerin (C_{21} H_{30} O_{13}.H_2O, amorphous, 6,8-di-O-acetyl barlerin hiside methyl ester), barlerin ($C_{19}H_{28}O_{12}$1.5 H_2O, amorphous, 8-0-acetyl shanzhiside methyl ester), and shanzhiside methyl esteer have been characterized. Flowers are reported to contain the flavone glycoside, scutellarein 7- neohesperidoside. The presence of B-sitosterol is reported in the plant.

The plant has antiseptic properties; its decoction is used as a wash in dropsy. The roots are used as a febrifuge. As a decoction they are employed as a mouthwash to relieve toothache, and as a paste they are applied over boils and glandular swellings. The dried bark is given in whooping-cough. Fresh juice of the bark is diaphoretic and expectorant, and is given in anasarca. The leaves and flowering tops are rich in soluble potassium salts, and are valued as a diuretic. The leaf juice mixed with honey or sugar is given in urinary and paralytic affections, and stomach disorders. The leaf juice is often applied to lacerated soles of feet in the wet season; with coconut oil it is applied on the face for pimples. The fresh leaves are crushed and tied on the wounds caused by sharp-edged tools. They are also used for rheumatic pains and itch[1,2,3,4,5].

MATERIAL AND METHODS

1. Procurement and authentication of Crude drugs:

The leaves of *Barleria prionitis* were collected in the month of September-October from Govt. garden, saket nagar, Bhopal. Plants were identified and authenticated in National Institute of Science Communication and Information Resources (NISCAIR/RHMD/Consult/2009-10/1359/161). The leaves were dried in shade and was powered moderately and passed through sieve No. 22/40.

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2.Evaluation Parameters:

(A) Pharmacognostic examination:

(i) Macroscopic examination: [6,7]

The leaf of *Barleria Prionitis* linn, dorsiventral, variable in size,6-9.5 cm long, 2.5 - 3.5 cm wide, simple, elliptic, acuminate, entire, acute, reticulate, unicostate, glabrous above, glabrous or pubescent beneath petiole short and the young stem grey, slightly four angled, usually with 3-4 divaricate spines at axil of leaf mature stem cylindrical with longitudinally arranged or scattered dot-like lenticels externally greyish to light brown a few mature stem slightly hollow.

(ii) Microscopic examination: [6,7]

Leaves of *Barleria prionitis*were washed and boil in water for 2 min until became soft enough to be cut using a sharp blade. Then transverse sections of leaves were observed under microscope.

lower epidermis; parenchyma many layered in upper surface and no of layers in lower surface towards proximal end, circular to polygonal and thin-walled; some contain raphides of calcium oxalate; vascular bundle semi-lunar, situated centrally in arenchymatous ground tissue; xylem vessels arranged in radial rows, protoxylem towards centre; two smaller vascular bundles present on either sides of central vascular bundle. Midrib - Single layered epidermis on both surfaces covered externally with thick cuticle. Collenchyma, parenchymatous cells shows single vascular bundle Lamina Single layered epidermis covered with thick cuticle on both surfaces, glandular (Figure 1)

(B) Micromeretic parameters: [8]

(i) Angle of repose:

Angle of repose is the maximum angle possible between the surface of a pile of the powder and the horizontal plane. (Table-1)

Procedure:

A glass funnel is held in place with a clamp on a ring support over a glass plate. Approximately 50 gm of powder is transferred in to the funnel by keeping the orifice of the funnel blocked by the thumb. As the thumb is removed, the lab-jack is adjusted so as to lower the plate and maintain about a 3 mm gap between the bottom of the funnel stem and the top of the powder pile. When the powder is emptied from the funnel, the angle of the heap to the horizontal plane is measured with the protractor and calculated by following formula.

$q = tan^{-1} h/r$

Where,

h = height of pile,

θ = angle of repose

r = radius of the base of the pile

(ii) Bulk Density:

Bulk density is the mass of powder divided by its bulk volume. A Powder (about 60 gm) was passed through a standard sieve no.20. A weighed amount (approx. 50gm) was introduced in to the bulk density apparatus, (aim is that to fill the measuring cylinder up to 75 ml) and the timer knob was set for 100 tapings. The volume occupied by the powder was noted. This final volume was the bulk volume. Then bulk density was calculated by using this equation. (Table-2)

$$\text{Bulk density } (\tilde{n}) = \frac{\text{Mass of powder}}{\text{Bulk volume}}$$

(iii) Tapped Density:

Tapped density was achieved mechanically by tapping a measuring cylinder containing a powder sample. After observing the initial volume, the cylinder was mechanically tapped, and volume readings were taken until little further volume change was observed. (Table-3)

$$\text{Tapped density } (\tilde{n}) = \frac{\text{Mass of powder}}{\text{Tapped volume}}$$

(C) Extraction:

The crude dried powdered drugs 50gm were kept for maceration in 200ml alcohol for 7 days. These drugs were remacerated and obtained extracts were further used for chemical evaluation.[9]

(D) Physical evaluation: [9]

(i) Determination of foreign matter:

About 10 gm of the sample was weighed and spread on a white tile uniformly without overlapping. Then the sample was inspected by means of 5x lens and the foreign organic matter was separated. After complete separation the matter was weighed and percentage w/w was determined. (Table-5)

(ii) Determination of solvent extractive value: [10]

Determination of water soluble extractive value:

Five gm of powdered drug was macerated with 100ml of water closed flask for 2hr and was occasionally shaked with 6hr time period and was allowed to stand for 18hr. After filtration the 25ml of the filtrate evaporated to dryness in a tarred flat bottomed shallow dish. Dried at 105°C and weighed. Percentage of water soluble extractive value was calculated with reference to the air dried drug. (Table-4)

Determination of alcohol soluble extractive value:

Alcohol is an ideal solvent for extraction of various chemicals like tannins, alkaloids, resins etc. Ethyl alcohol (95% v/v) was used for determination of alcohol soluble extractive.

Five gm of powdered drug was macerated with 100ml of ethanol closed flask for 24hr and was occasionally shaked with 6hr time period and was allowed to stand for 18hr. After filtration the 25ml of the filtrate evaporated to dryness in a tared flat bottomed shallow dish. Dried at 105°C and weighed. Percentage of ethanol soluble extractive value was calculated with reference to the air dried drug. (Table-4)

(iii) Determination of Moisture Content: [9,10]

The percentage of active constituents in crude drug is mentioned on air dried bases. Hence, the moisture content of the crude drugs should be determined and should also be controlled. The moisture content should be minimized in order to prevent decomposition of crude drugs either due to chemical changes or microbial contamination.

Procedure : The powdered sample of leaves of *Barleria Prionitis*weighed 5gm accurately and kept in IR moisture balance. The loss in wt. was recorded as percentage (%) moisture with respect to air-dried sample of crude drug. (Table-4)

(iv) Determination of Ash value: [10,11]

The residue remaining after incineration is the ash content of the drug, which simply represents inorganic salts, naturally occurring in drugs or adhering to it or deliberately added to it as a form of adulteration. Many a time the crude drugs are admixed with various mineral substances like sand, soil, calcium oxalate, chalk powder or other drugs with different inorganic content. Ash value is a creation to judge the purity of crude drugs. Generally either total ash value or acid-insoluble ash value or both is determined. Total ash usually consists of phosphates, silicates and silica. On the other hand, acid-insoluble ash, which is a part of total ash insoluble in dilute hydrochloric acid, contains adhering dirt and sand.

Determination of total ash:

Total ash was determined by weighing 2 gm of the air dried crude drug in the tared platinum or silica dish and incinerated at a temperature not exceeding 450°C until free from carbon and then was cooled and weighed. (Table-5)

Determination of acid insoluble ash:

The ash obtained from the previous process was boiled with 25ml of 2M HCI for 5 min. and the insoluble matter was collected on ash-less filter paper and was washed with hot water, ignited, cooled in a dessicator and weighed. Percentage of acid insoluble ash was calculated with reference to the air dried drug. (Table-5)

Determination of water soluble ash:

The ash was boiled with 25ml of water for 5 min. and the insoluble matter was collected on ash-less filter paper and was washed with hot water, ignited for 15min. at a temperature not exceeding 450°C. The weight of the insoluble matter was subtracted from the weight of the ash and this represents the water soluble ash. Percentage of water soluble ash was calculated with reference to the air dried drug. (Table-5)

E. Qualitative Phytochemical analysis:

The extracts obtained were subjected to various qualitative tests to reveal the presence or absence of common phytopharmaceuticals by using standard tests. [6,7,12] (Table-6)

RESULT

Table-1 Angle of repose

Drug	Height of pile(cm)	Diameter of the base of the pile (cm)	Radius of the base of the pile (cm)	Angle of) repose (q)
Barleia prionitis L	4.8	13	6.5	36.12

Table-2 Bulk Density (g/cm^3)

Drug	I	II	III	Average
Barleia prionitis L	0.33	0.33	0.33	0.33

Table-3 Tapped Density (g/cm^3)

Drug	I	II	III	Average
Barleia prionitis L	0.34	0.34	0.35	0.34

Table-4 Solvent Extractive Value

Name of the drug	Water soluble Extractive value %w/w	Alcohol soluble Extractive value %w/w	Chloroform Soluble Extractive value %w/w	Moisture content (% w/w)
Barleia prionitis L	28..88	10.45	4.68	3.2

Table-5 Physico-chemical Characteristics

Name Of The Drug	Foreign Organic Matter (%)	Total Ash Value (%)	Acid Insoluble Ash Value(%)	Water Soluble Ash Value(%)
Barleia prionitis L	0.8	6.1	.90	.90

Table-6 Qualitative Phytochemical Test

S.No.	Test for	Result
1.	Alkaloids	+
2.	Carbohydrates	–
3.	Glycosides	+
4.	Volatile oil	-
5.	Proteins	+
6.	Terpenoids	_
7.	Gums & resins	–
8.	Tannins	+
9.	Flavanoids	+

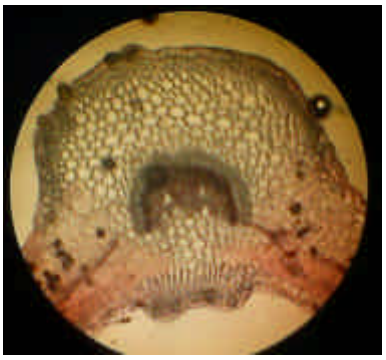

Figure-1 *Transverse Section. of Barleria Prionitis leaves*

DISCUSSION

Macroscopic study reveals that the *Barleria Prionitis*leaves were dorsiventral, variable in size,6-9.5 cm long, 2.5 - 3.5 cm wide, simple, elliptic, acuminate, entire, acute, reticulate, unicostate, glabrous above, glabrous or pubescent beneath petiole short and the young stem grey, slightly four angled, usually with 3-4 divaricate spines at axil of leaf mature stem cylindrical with longitudinally arranged or scattered dot-like lenticels externally greyish to light brown a few mature stem slightly hollow.

Under microscopic study we found lower epidermis; parenchyma many layered in upper surface and no. of layers in lower surface towards proximal end, circular to polygonal and thin-walled; some contain raphides of calcium oxalate; vascular bundle semilunar, situated centrally in arenchymatous ground tissue; xylem vessels arranged in radial rows, protoxylem towards centre; two smaller vascular bundles present on either sides of central vascular bundle. Midrib - Single layered epidermis on both surfaces covered externally with thick cuticle. Collenchyma, parenchymatous cells shows single vascular bundle Lamina Single layered epidermis covered with thick cuticle on both surfaces, glandular

Micromeretic parameters such as angle of repose, bulk density and tapped density were found to be 36.12, 0.33, and 0.34 respectively. Water, alcohol and chloroform soluble extractive value were found to be 28.88 % (w/w), 10.45 % (w/w) and 4.68 % (w/w) respectively. Moisture content and foreign organic matter were 4.2 % (w/w) and 1.8 % respectively. Total ash value, acid insoluble ash and water soluble ash were found to be 6.1%, 9%, and 9% respectively. Qualitative phytochemical tests showed the presence of alkaloids, Flavanoids & Tannins in ethanolic extract of *Barleria Prionitis* leaves.

CONCLUSION

From the above studies it can be concluded that the various parameters such as pharmacognostical, phytochemical and micromeritics parameters of the leaves of *Barleria Prionitis* may be utilized for its identification and differentiation from other species. Phytochemical investigation reveals the presence of alkaloids and Flavonoid's, tannins in etanolic extract of *Barleria Prionitis* leaves. Due to presence of these compounds in the leaves of *Barleria Prionitis*, it may have good antioxidant activity, anti-inflammatory activity, Gastric cytoprotective activity and diuretic activity etc. Thus the *Barleria Prionitis* leaves may be a good choice for the futuristic research on such activities.

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