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PHYSICOCHEMICAL AND PHYTOCHEMICAL ANALYSIS OF *BOERHAVIA DIFFUSA* LINN (PUNARNAWA) ROOT

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ABSTRACT

Boerhaavia diffusa Linn. (family- Nyctaginaceae), commonly known as 'Punarnava' in the Indian system of medicine. It is found throughout India. Root and aerial parts of 'Punarnava were used in of diabetes, digestive system, heart diseases anemia, and respiratory distress, liver problems. The present paper provides a detail account of the *Boerhaavia diffusa* Linn. root. The study includes macroscopic, HPTLC fingerprinting, preliminary phytochemical screening, and physicochemical parameters. The information generated will be useful for proper identification and authentication of *Boerhaavia diffusa* Linn. root.

KEYWORDS: Boerhaavia diffusa Linn., Pharmacognostic evaluation, Preliminary phytochemical screening, Physico chemical analysis, HPTLC fingerprinting.

INTRODUCTION

Boerhavia diffusa Linn. is commonly known as Punarnava. It is used to relieve pain and the leaves are used as a green vegetable in numerous parts of India. It is found in Australia, China, Egypt, Pakistan, Sudan, Sri Lanka, South Africa, USA and in several countries of the Middle East. It is a perennial, spreading hogweed, commonly occurring abundantly in waste places, ditches and marshy places during rains. The plant is also cultivated to some extent in West Bengal.[1,2] It grows well on wastelands and in fields after the rainy season.[3] The whole plant and preferably the roots are effectively used to cure several diseases including Jaundice.[4] Punarnava corrects the digestive system, alleviates fluid retention and very useful in managing heart diseases. It is also used to treat the anemia, hernia and respiratory distress, liver problems, managing lipids and cholesterol in healthy limits[5] Despite the numerous

medicinal uses attributed to this plant, there are no pharmacognostical studies on the root of this plant have so far been carried out. Hence, the present work deals with the physicochemical constants, preliminary phytochemical screening and HPTLC fingerprint profile of Punarnava root which could serve as a valuable source of information and provide suitable standards for the further identification of this plant.

MATERIALS AND METHODS

Collection of specimens

The fresh plant root of *Punarnawa was* collected from the Chitrakoot forest of Satna district (M.P.) in the month of March 2016. Fresh material was used for anatomical studies whereas shade dried material was powdered in electric grinder for physico-chemical, phytochemical and HPTLC studies.

Macroscopic study

Macroscopic or organoleptic characters like appearance, colour, odour and taste were evaluated.[6]

Fluorescence study

Fluorescence study was carried out with different chemicals and colour observed with the help of UV Spectrophotometer.[7]

Physico-chemical study

Physico-chemical parameters such as moisture content (loss on drying at 105^0C), water soluble extractive value, alcohol soluble extractive value, total ash value, acid insoluble ash value and water soluble ash were calculated.[8,9]

Preliminary phytochemical screening

Preliminary tests were carried out on ethanolic and water extract for the presence\absence of phyto-constituents like alkaloids, flavanoids, tannins, resins, carbohydrates, proteins and saponins.[10,11]

High Performance Thin Layer Chromatography (HPTLC)

For HPTLC, the 2 gm root powder was extracted with 50 ml of ethanol overnight, filtered and concentrated. It was applied by spotting extracted sample on pre-coated silica-gel aluminium plate 60 F_{254} (5x10 cm with 0.2 mm layer thickness Merk Germany) using Camag Linomat -5 sample applicator and a 100 µl Hamilton syringe. The samples, in the form of

bands of length 6 mm, were spotted 15 mm from the bottom, 15 mm from left margin the plate and 10 mm part. Plates were developed using mobile phase consisting of *Toluene: Ethyl acetate (7:3* v\v). Linear ascending development was carried out in 10x10cm twin through glass chamber equilibrated with mobile phase. The optimized chamber saturation time for mobile phase was 30 min. at room temperature. The length of chromatogram run was 8 cm. 20 ml of the mobile phase. Subsequent to the development, TLC plates was dried with the help of Hot Air Oven. The peak area for samples and standard were recorded with Camera photo documentation system Camag Reprostar 3. Visualization of spots were made before and after derivatization (with 5% *Methanolic- sulphuric* reagent) at 254nm, 366nm and day light with Win cat software and R_f values noted.[12-14]

RESULTS AND DISCUSSION

Macroscopy

Punarnawa root colour is brown, taste slightly bitter and odour characteristics.

Physico-chemical analysis

The physico-chemical parameters such as extractive values are useful for the determination of exhausted or adulterated drug; ash values of the drug gave an idea of the earthy matter or the inorganic composition and other impurities present along with the drug. Physico-chemical results of the drug are given in (Table1).

Florescence study

Florescence study were done and results are given in table 2.

Preliminary phytochemical studied

Qualitative phyto-constituents were screened in the extracts taken in water and ethyl alcohol. The screening exhibited presence of saponin, alkaloids, tannin and resin.

HPTLC finger print profile

High performance thin layer chromatography (HPTLC) study of the ethanolic extract two spots of the sample extracts applied in the TLC plate. Major spots R_f values with colour were recorded under 366nm, after derivatization 366nm and UV light. Chromatogram profile and R_f values are given (Fig.1 - 2 & Table 3).

Table 1: Physico-chemical analysis of Punarnawa root.

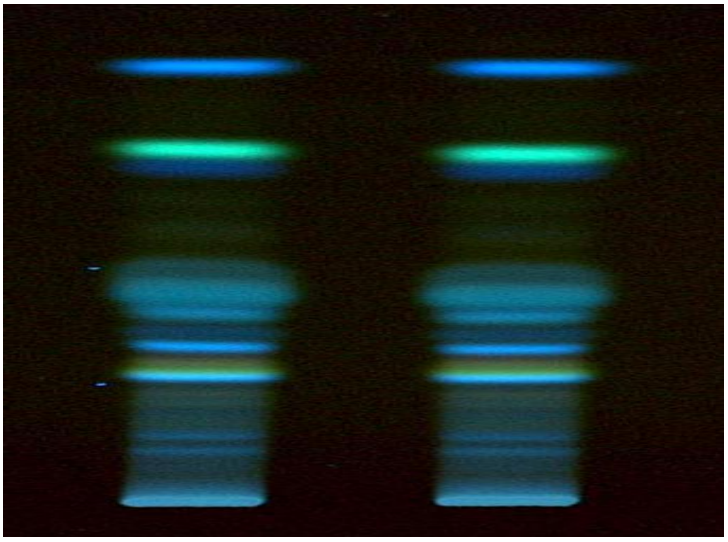
S. N0.	Parameters	Result
1	Foreign matter	2.15%
2	Loss on drying(at 105^{o}C)	5.50%
3	Alcohol soluble extractive value	14.22%
4	Water soluble extractive value	23.60%
5	Total ash value	10.12%
6	Acid in soluble ash value	1.08%

Table 2: Fluorescence study of Punarnawa root.

S.No.	Powder + reagent	Observation at day light	Observation at 366nm
1	Powder a sit +P	Brown colour	Dark cream
2	1 N HCl + P	Brown	Dark green
3	1 NaoH (methanol) + P	Green	Light green
4	1 NaoH +P	Dark brown	Dark green
5	50% KOH + P	Dark brown	Green
6	50% H2SO4 + P	Dark brown	Dark green
7	Con. H2SO4 + P	Black	Black
8	50% HNO3 +P	Brown	Black
9	Con. HNO3 + P	Brownish red	Black
10	Glacial acetic acid	Brownish green	Green
11	Iodine water + P	Brown	Black
12	50% HCl + P	Brown	Green
13	Con. H Cl+ P	Brownish black	Green
14	Picric acid +P	Brown	Light green
15	Acetone + P	Brown	Whitish green
16	50% FeCl3 + P	Dark green	Black
17	50% Ammonia +P	Brown	Green

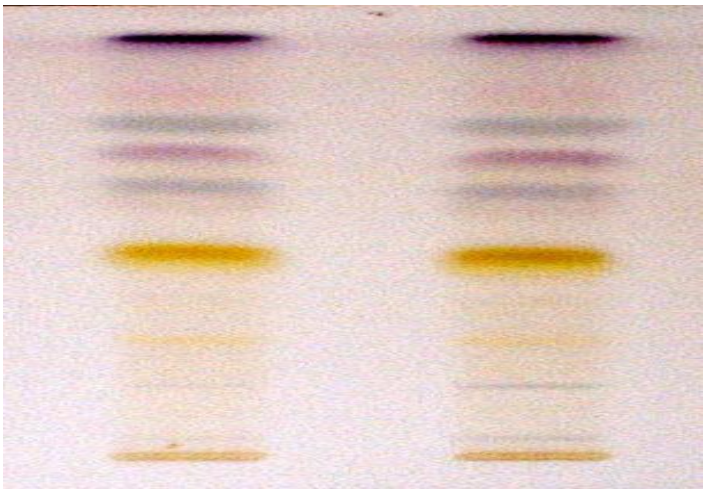
Table 3: Rf values of HPTLC fingerprint profile of Punarnawa root.

Rf values	At 366nm Before derivatization		At visible light	
	Test solution S1	Test solution S2	Test solution S1	Test solution S2
R_f 1	0.12(sky blue)	0.12(sky blue)	0.08(light brown)	0.08(light brown)
R_f 2	0.14(sky blue)	0.14(sky blue)	0.24 (light brown)	0.24 (light brown)
R_f 3	0.22(sku blue)	0.22(sku blue)	0.30 (light yellow)	0.30 (light yellow)
R_f 4	0.24(yellowish green)	0.24(yellowishgreen)	0.40(pinkish brown)	0.40(pinkish brown)
R_f 5	0.30 (sky blue)	0.30 (sky blue)	0.52(brown)	0.52(brown)
R_f 6	0.36(sky blue)	0.36(sky blue)	0.60 (yellow)	0.60 (yellow)
R_f 7	0.40 (sky blue)	0.40 (sky blue)	0.70(brown)	0.70(brown)
R_f 8	0.58(sky blue)	0.58(sky blue)	0.80(black)	0.80(black)
R_f 9	0.60 (whitish yellow)	0.60 (whitish yellow)	-	-
R_f 10	0.80(sky blue)	0.80(sky blue)	-	-



S_1 **S_2**

Fig. 1: HPTLC fingerprint at 366nm before derivatization.



S_1 **S_2**

Fig. 2: HPTLC fingerprint after derivatization UV light.

CONCLUSION

The physicochemical data and and phytochemical values reported in this work may play a major role in setting some diagnostic characters for identification of the plant. With the help of this referential information, a researcher can easily reject the fake and adulterated plant products which are deviated from the above mentioned characters and select the correct herbal specimen for further investigations.

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