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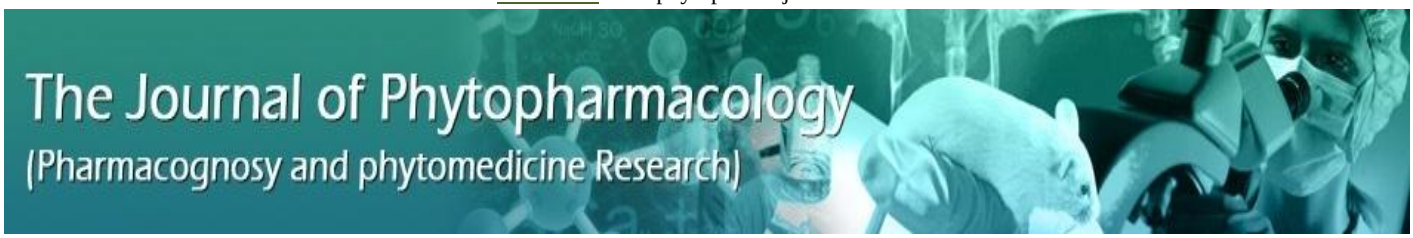

Research Article

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Standardization of Haridradi churna- physicochemical assay and HPTLC profile

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ABSTRACT

Haridradi churna, a polyherbal preparation containing Haridra (*Curcuma longa* Linn.), Devadaru (*Cedrus deodar* Roxb), Lodhra (*Symplocos racemosa* Roxb), Priyangu (*Callicarpa macrophylla* Vahl) and Yastimadhu (*Glycyrrhiza glabra* Linn.) has been mentioned in Ayurveda texts for umbilical cord care in new born baby. Since the therapeutic values and efficacy of the herbals depend on many factors such as geographical variations, seasons of collection and method of preparation, a physicochemical assay and HPTLC analysis of the above formulation has been taken up in the present study. Standard protocols for AYUSH drugs have been followed in the study. The HPTLC profile of the alcohol extract of the drug revealed the presence of thirteen photochemical with R_f values ranging from 0.04 to 0.95. The results are useful in quality control and standardization of Haridradi churna formulation.

Keywords: Haridradi churna, cord care, physicochemical assay, HPTLC profile.

INTRODUCTION

There has been a continued increase in demand for Ayurvedic medicinal preparations for the treatment of various types of human ailments. This appears to be due to their better tolerance and negligible adverse side reactions[1]. WHO has considered phytotherapy in its health programmes because these drugs are safe, cost effective and most importantly people have faith in them. The global increase in demand for herbal drugs has also created the danger of drug adulteration and contamination[2]. This emphasizes the need for the rigorous quality control and standardization of herbal preparations through appropriate scientific testing and evaluation methods[3].

Haridradi churna is a polyherbal preparation containing Haridra (*Curcuma longa* Linn.), Devadaru (*Cedrus deodar* Roxb), Lodhra (*Symplocos racemosa* Roxb), Priyangu (*Callicarpa macrophylla* Vahl) and Yastimadhu (*Glycyrrhiza glabra* Linn.). Its therapeutic values and preparation have been highlighted in Ayurvedic texts[4].

The present study is aimed at physicochemical assay and HPTLC finger printing of Haridradi churna as a part of standardization of the formulation.

MATERIALS AND METHODS

The raw materials for the formulation- Haridra (*Curcuma longa* Linn.), Devadaru (Cedrus deodar Roxb), Lodhra (*Symplocos racemosa* Roxb), Priyangu (*Callicarpa macrophylla* Vahl) and Yastimadhu (*Glycyrrhiza glabra* Linn.) were procured from SDM Ayurvedic Pharmacy, Kuthpady, Udupi, Karnataka state, India and got authenticated at SDM Center for Research in Ayurveda and Allied Sciences, Kuthpady, Udupi, Karnataka state, India with following standard methods.

Preparation of Haridradi churna

The dried individual raw materials were powdered separately and passed through sieve number 85 to obtain fine powder. They were mixed in equal proportions to obtain a homogeneous mixture and packed in air-tight containers.

Physicochemical analysis

Physicochemical studies such as loss on drying at 105°C, total ash, water soluble ash, acid insoluble ash, alcohol soluble extractive value, water soluble extractive values were carried out according to official methods[5,6,7].The HPTLC studies were carried out following the methods of Harborne and Wagner *et al*.,[8, 9].

Loss on drying at 105°C

10 grams of the churna was placed in a tarred evaporating dish. It was dried at 105˚C for 5 hours in a hot-air oven, cooled to room temperature in desiccators and weighed. The drying, cooling and weighing was continued until difference between two successive weights was not more than 0.01. The percentage of moisture was calculated with reference to the weight of the sample.

Determination of Total Ash

2 grams of the sample was incinerated in a clean and pre-weighed tarred platinum crucible at a temperature not less than 450°C until carbon free ash was obtained. Percentage ash was calculated with reference to the weight of the sample.

Acid insoluble ash

To the crucible containing total ash 25ml of dilute hydrochloric acid was added, boiled and cooled. The insoluble matter was filtered using an ash less filter paper (Whatman 41), washed with hot water till free from acid. The filter paper along with the insoluble matter was taken in the same crucible, dried in a hot-air oven and ignited to constant weight. The percentage of acid-insoluble ash was calculated with reference to the air-dried drug.

Water Soluble Ash

The ash was boiled with 25 ml of water for 5 minutes. The insoluble matter was collected on an ash less filter paper, washed with hot water, ignited for 15 minutes at a temperature not exceeding 450C, cooled and weighed. The difference in weight of the insoluble matter and the weight of total ash represents the water-soluble ash. The percentage of water soluble ash with reference to the air-dried drug was calculated.

Alcohol Soluble Extractive Value

5grams of the powdered air-dried drug was taken in a glass Stoppard flask. 100ml of distilled alcohol (approximately 95%) was added, shaken frequently for 6 hours and then allowed to stand for 18 hours. It was filtered rapidly taking precautions against loss of solvent.25ml of the filtrate was evaporated to dryness in a tarred flat-bottomed shallow dish at 105°C to constant weight. The percentage of alcohol-soluble extractive was calculated with reference to the weight of air dried drug.

Water soluble extractive

4 grams of the sample was taken in a glass Stoppard flask with 100 ml of distilled water and shaken occasionally for 6 hours. It was allowed to stand for 18 hours and filtered.25ml of the filtrate was pipetted out into a pre-weighed 100 ml beaker and evaporated to dryness on a water bath. It was further dried in a hot-air oven at 105°C for 6 hours, cooled in a desiccator and weighed. The percentage of water -soluble extractive was calculated with reference to the weight of air dried drug.

Determination of pH value

The pH of the aqueous solution of the sample was determined with the help of pH meter and glass electrode system at laboratory temperature.

HPTLC finger printing

HPTLC system from M/s CAMAG, Muttenz, Switzerland consisting of Linomat 5 sample applicator, twin-trough chamber, TLC visualiser, TLC scanner 3 linked to CATS software was used.

One gram of the powdered sample was dissolved in 10ml ethanol and kept for cold percolation for 24h and filtered. 3, 6 and 9µl of the above sample was applied on a pre-coated silica gel F254 (E. Merk) on aluminium plate to a band width of 7 mm using Linomat 5 TLC applicator. The plate was developed in Toluene: Ethyl acetate, formic acid (5.0: 1.5: 0.5) solvent system. The developed plates were dried at 105°C and visualized with UV 254nm, 366 nm and then derivatised with vanillin sulphuric acid reagent and scanned under white light. The densitometric scans with the useful data were recorded.

RESULTS AND DISCUSSION

The physicochemical parameters of Haridradi churna are presented in table 1.

Table 1: Results of physicochemical parameters ofHaridradi churna

Parameter	Resultsn = 3%w/w
Loss on drying	10.80
Total Ash	6.68
Acid Insoluble Ash	0.80
Water soluble Ash	2.50
Alcohol soluble extractive value	9.96,
Water soluble extractive value	15.97
pH	4.7

The total ash content represents inorganic salts naturally occurring/adhering in raw materials or added as adulterants. This parameter serves to detect contamination and adulteration by sand or earth. The low ash content 6.68%w/w of Haridradi churna indicated purity of the formulation.Extractive values indicate the active component present in the plant material. Water soluble and alcohol soluble extractives is used as a means of evaluating crude drug. Fairly high values of water soluble extractive (15.97%w/w) and alcohol soluble extractive (9.96%w/w) confirm the presence of higher amounts of the active principles in the formulation.

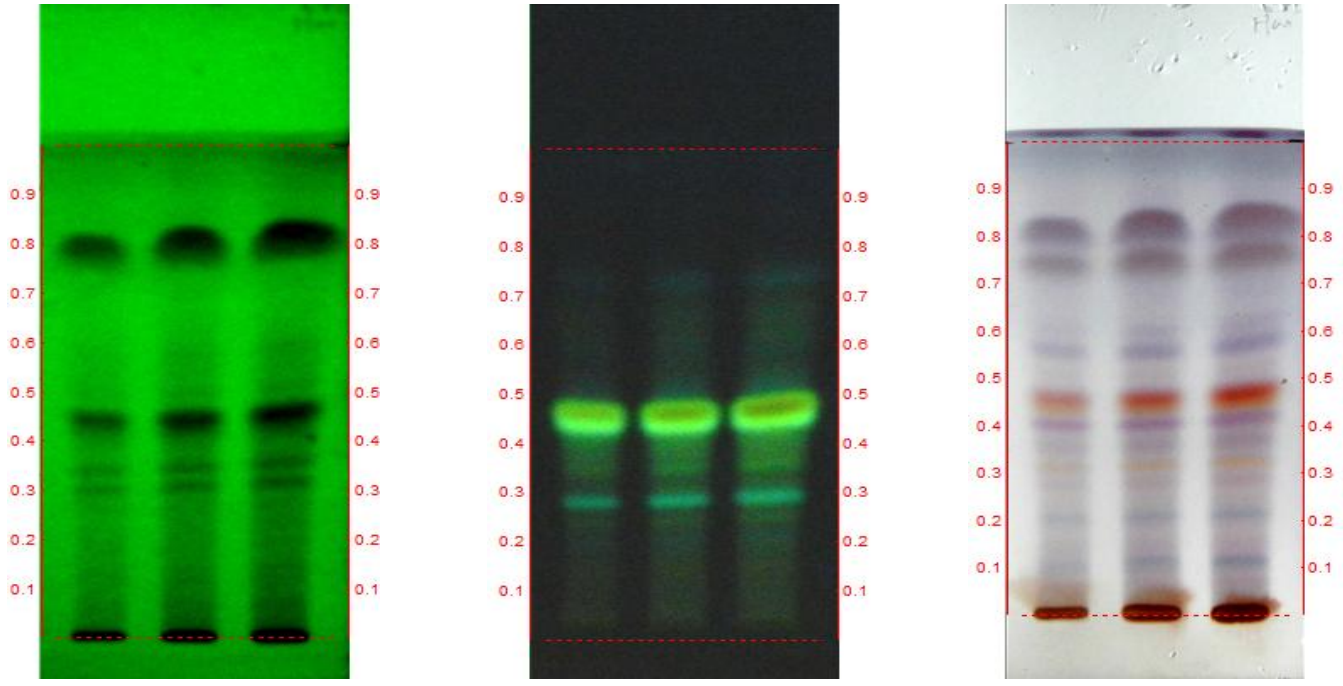
Dryness of the sample is important in size reduction, preservation and preventing hydrolytic degradation of active components of herbal formulations. Higher moisture content promotes growth of microbes and hence spoilage of formulations[10]. The moisture content was found to be low (10.8% w/w) in the Haridradi churna.

pH value is a measure ofacidity or alkalinity of the aqueous solution of the sample.It is important from the standpoint of physiological suitability. The pH value 4.7 suggested a slight acidic nature of the formulation.

HPTLC profile

The HPTLC finger print profile (densitometric scan) of ethanolic extract of Haridradi churna at UV 254nm, 366nm and post-derivatisation with white light is presented in figures 1 to 4. The densitometric scan at UV 254nm revealed the presence of thirteen phytochemical components (Fig. 2) with R_f values ranging from 0.04 to 0.95. It is clear from the chromatogram that the three components with R_f values 0.04, 0.54 and 0.95 are present in higher concentrations with peak areas 17.69%, 36.57% and 18.83 %, respectively. However, the scan at 366nm revealed the presence of twelve components (Fig.3) with R_f values ranging from 0.04 to 0.97 and higher peak areas for the components with $R_f$values 0.04 (19.92%), 0.34 (10.98%), 0.45 (11.24%) & 0.54 (36.57%). The postderivatised chromatogram showed eleven phytochemical components (Fig.4) with R_f values ranging from 0.05 to 0.96.

The HPTLC finger prints are useful to verify the quality and purity of drug in compound formulations[11].



At 254nm **At 366nm** **Under white light post derivatisation**

Figure1: HPTLC finger print profile of alcoholic extract of Haridradi churna.
Solvent system- Toluene: Ethyl acetate: Formic acid (5.0: 1.5: 0.5); Track 1: 3µl, Track 2: 6µl, Track 3: 9µl.

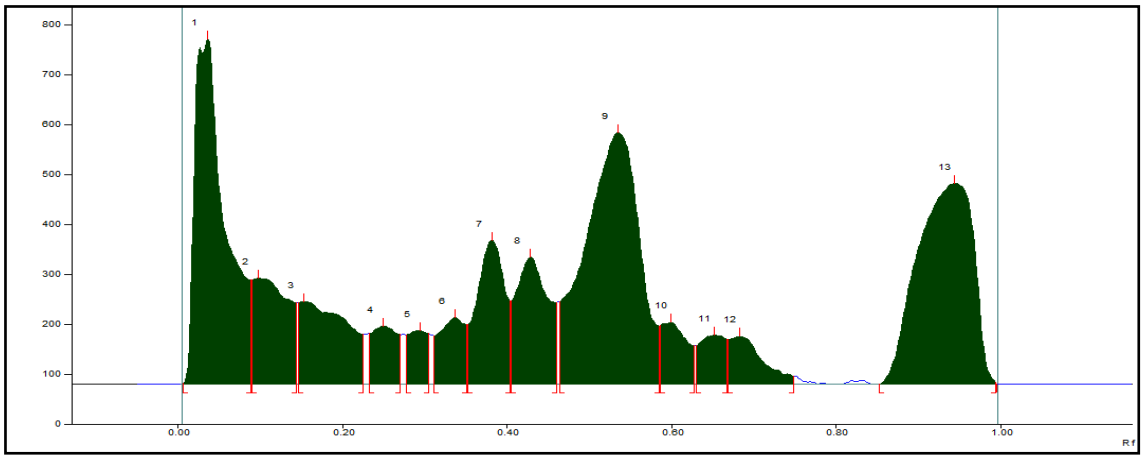


Figure 2: Densitometric scan at 254nm

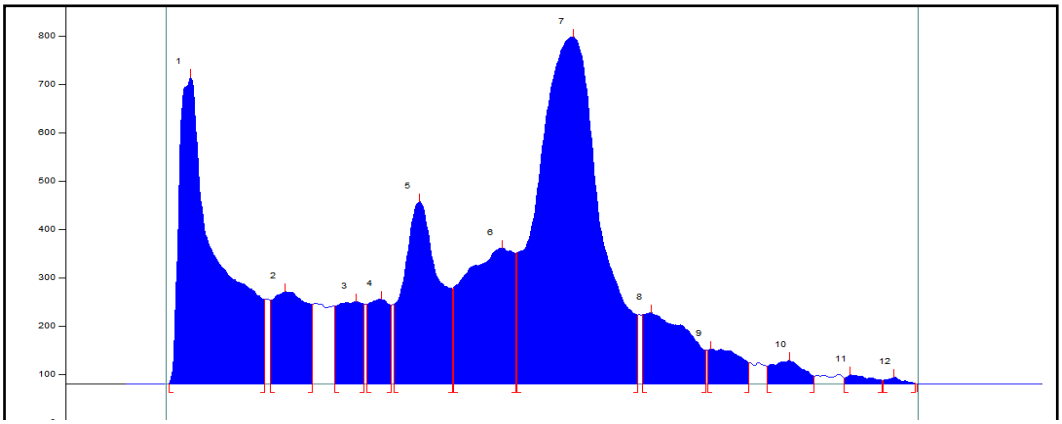


Figure 3: Densitometric scan at 366nm

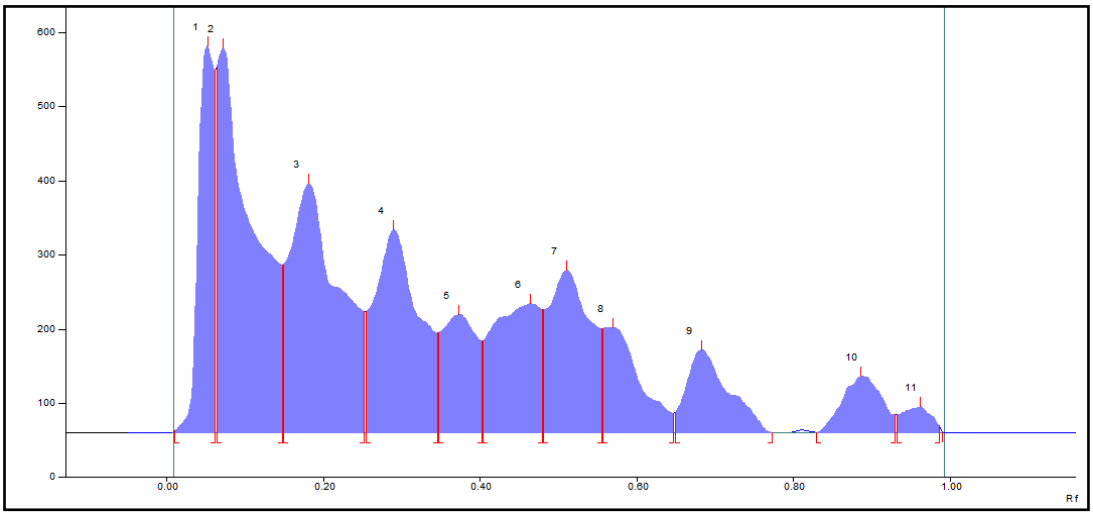


Figure 4: Densitometric scan after postderivatization

CONCLUSION

Characterization of the Haridradi churna has been done as per pharmacopoeial methodology. The set of data obtained in the present investigation can serve as standard for the identification of churna. The results will be helpful in sustaining and reproducibility of drug as well as to ensure the quality and safety of drug.

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