# Pharmacognostical, physicochemical and high performance thin layer chromatography evaluation of *Dhatryadi kwatha*

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#### Abstract

**Background:** *Dhatryadi kwatha* is mentioned in Ayurvedic classics as a therapeutic formulation to treat *Shwitra* (Vitiligo). Vitiligo affects 1% of the population worldwide, but management is until unsatisfactory. *Dhatryadi kwatha* contains *Dhatri (Emblica officinalis* Gaertn.), *Khadira (Acacia catechu* Willd.), and *Bakuchi (Psoralea corylifolia* Linn.). *Dhatri* and *Khadira* both have *Kushthaghna* property. The skin diseases are considered in the umbrella of *Kushtha*. *Bakuchi* has also *Kushthaghna* property. Thus, it is more effective in Vitiligo internally. **Materials and Methods:** *Dhatryadi kwatha* powder was evaluated for their pharmacognostic and pharmaceutical analysis. **Results:** Microscopic characters were found of *Amalaki, Khadira*, and *Bakuchi*. Results obtained in pharmaceutical parameters of *Dhatryadi kwatha* powder like loss on drying 15.19%, ash value 8.48%, alcohol soluble extract 58.6% w/v, etc., are within limit mentioned by Ayurvedic Pharmacopoeia of India. High performance thin layer chromatography profile of *Dhatryadi kwatha* powder showed similarities in number of spots. **Conclusion:** From the study, data developed can be espoused for laying down the standards for *Dhatryadi kwatha*.

Key words: Dhatryadi kwatha, high performance thin layer chromatography, pharmaceutics, pharmacognocy, Shwitra, Vitiligo

# **INTRODUCTION**

hatryadi kwatha comprising Amalaki, Khadira, and Bakuchi was first explained in Chakradatta for curing of *Shwitra* (Vitiligo).<sup>[1]</sup> During the last decades, herbal medicines pointed out in Ayurveda are getting gratitude globally. Maintaining the quality standard of a polyherbal formulation is a challenging task. Available data concerning the scientific evaluation of Dhatryadi kwatha are none. Quality control for safety and efficacy of herbal products is of paramount importance.<sup>[2,3]</sup> With the help of identity, purity, content, and other chemical, physical, or biological properties, or by the manufacturing processes quality can be defined as the status of a drug. The analytical techniques have always been cited to understand the quality of the outcome in Ayurveda. It describes different qualitative parameters to critic genuine plant identification, preparations and having scientific evidence; they are not competent to provide quantitative information. Using the modern techniques, qualitative and quantitative analysis of drugs and instruments of the science is of absolute importance to rationalize their acceptability in the modern system of medicine.

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**Received:** 11-12-2016 **Revised:** 30-12-2016 **Accepted:** 07-01-2017 The different chromatographic analysis is routinely used and plays an important role in the quality control of complex herbal medicines. High performance thin layer chromatography (HPTLC) can provide an electronic image of the chromatographic fingerprint and a densitogram to detect the presence of marker compounds in a plant sample. The advantage of HPTLC in the analytical testing of herbal products is that it provides positive identification as well as visualization of the separated fractions of the sample component and helps in quantitative, qualitative analysis with the same system.

Dhatri stands synonym of Amalaki (Emblica officinalis), Khadira (Acasia catechu), and Bakuchi (Psoralea corylifolia). Dried Fruits, heartwood, and seeds were used respectively of these herbs have high medicinal value. Dhatryadi kwatha is used as drug of choice for Shwitra (Vitiligo). Hence, this study is anticipated to evaluate Dhatryadi kwatha powder through pharmacognostic, physicochemical, and HPTLC analysis.

#### Aim

The aim of this study was to authenticate the *Dhatryadi kwatha* as per pharmacopeial (Ayurvedic Formulary of India and Ayurvedic Pharmacopoeia of India) method and to evaluate the quality of the drug.

## **MATERIALS AND METHODS**

#### **Collection and Preparation of the Drug**

Fruits of *Amalaki*, heartwood of *Khadira*, and seeds of *Bakuchi* were collected from the pharmacy of Institute for Postgraduate Teaching and Research in Ayurveda, Jamnagar. The obtained drugs were shade dried, equally amount had taken and made into a coarse powder with the help of mechanical grinder. Ingredients of *Dhatryadi kwatha* are summarized at Table 1.

#### **Organoleptic Evaluation**

Various parameters of the material such as color, odor, touch, and taste of the *kwatha* powder were observed and recorded [Table 2].<sup>[4]</sup>

#### **Microscopic Evaluation**

Microscopic examination of material powder was carried out with and without staining, by powder microscopy to determine the chemical nature and microphotographs were taken using Carl Zeiss binocular microscope.<sup>[5]</sup>

#### **Physicochemical Analysis**

Physicochemical analyses were performed by following the parameters. Physicochemical analysis such as loss on drying at 110°C,<sup>[6]</sup> pH value,<sup>[7]</sup> ash value,<sup>[8]</sup> water-soluble extractive,<sup>[9]</sup> and methanol soluble extractive<sup>[10]</sup> were recorded.

#### **Preliminary Phytochemical Investigation**

Preliminary phytochemical investigations are carried out by following standard procedure of API.<sup>[11]</sup>

#### HPTLC

HPTLC was performed as per the guidelines provided by API.<sup>[12]</sup> A CAMAG (Switzerland) HPTLC system equipped with a sample applicator Linomat V was used for application of samples. The methanol extract of *kwatha* powder was used for spotting. Toluene:ethyl acetate:acetic acid (7:2:1 v/v) was selected as the solvent system. CAMAG TLC Scanner 3, Reprostar and Wincats 1.3.4 were used for scanning the plates. CAMAG twin trough glass chamber was used for developing the plates. The developed plate was visualized under visible daylight, short ultraviolet (UV) (254 nm), long UV (366 nm) and after spraying with vanillin-sulfuric acid reagent and again observed in daylight. The reference values were recorded.

#### Instrumental Conditions

Application mode: Camag Linomat V, development chamber: Camag twin trough chamber, plate: Precoated Silica Gel GF254 plate, chamber saturation: 30 min, development time: 30 min, development distance: 10 cm, scanner: Camag scanner III, detection: Deuterium lamp and mercury lamp, and data System: Win CATS software.

Table 1: Ingredients of Dhatryadi kwatha				
Sanskrit name	Latin name	Parts used	Quantity	
Dhatri (Amalaki)	E. officinalis (Gaertn.)	Phala (fruits)	1 part	
Khadira	A. catechu (Wild.)	Heartwood	1 part	
Bakuchi	P. corylifolia (Linn.)	<i>Beeja</i> (seeds)	1 part	

E. officinalis: Emblica officinalis, A. catechu: Acasia catechu, P. corylifolia: Psoralea corylifolia

# **OBSERVATIONS AND RESULTS**

#### Pharmacognostic Study

Microscopic powder characters of *Amalaki* were found such as mesocarp cells, epicarp cells, fibers, and groups of scleroids. Microscopic powder character of heartwood of *Khadira* was found that silica deposition. Microscopic powder characters of seeds of *Bakuchi* were found such as scleroids, epicarp cells, pitted vessels, and oleoresin contents which are depicted in Table 3 and Figure 1.

#### **Analytical Study**

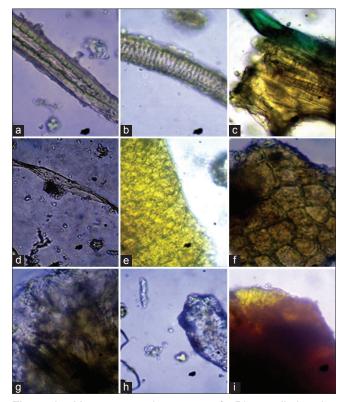
Results of the analytical study of *Dhatryadi kwatha* powder are as follows.

#### **Physicochemical Constants**

The results are depicted in Table 4.

#### HPTLC

In HPTLC, in short UV-254 nm, maximum 8 spots were observed in *Dhatryadi kwatha*. Similarly, in long



**Figure 1:** Microscopic characters of *Dhatryadi kwatha:* (a) Scleroids of *Bakuchi*, (b) pitted vessels of *Bakuchi*, (c) Group of scleroids of *Amalaki*, (d) fibers of *Amalaki*, (e) epicarp cells of *Bakuchi*, (f) epicarp cells of *Amalaki*, (g) mesocarp cells of *Amalaki*, (h) silica deposition of *Khadirasara*, (i) oleoresin content of *Bakuchi* 

UV-366 nm, maximum 8 spots were observed [Table 5 and Figure 2].

Nature of adsorbed components, if with different polarity, formerly total number of components and respective reference values also differs. In short, nature of different matrix modulates both the studied parameters.

#### **DISCUSSION AND CONCLUSION**

Results obtained in physicochemical parameters of *Dhatryadi kwatha* are within limit mentioned by Ayurvedic Pharmacopoeia of India. HPTLC profile of *Dhatryadi kwatha* showed similar in number of spots. This profile can be used

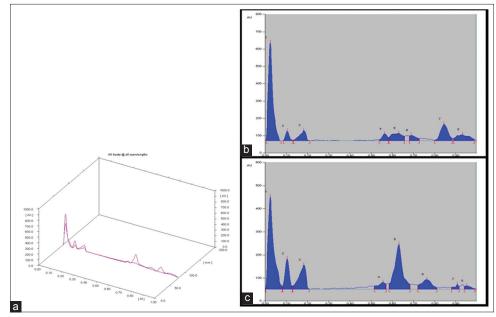
Table 2: Organoleptic characters of Dhatryadikwatha		
Organoleptic characters	Results	
Color	Brownish muddy	
Odor	Aromatic	
Taste	Bitter	
Touch	Rough	
Appearance	Powder	

Table 3: Microscopic characters of Dhatryadi kwatha		
Name of drug	Characters found	
Amalaki	Mesocarp cells, Epicarp cells, fibers, and groups of scleroids	
Khadira	Silica deposition	
Bakuchi	Epicarp cells, pitted vessels, and scleroids	

# Table 4: Physicochemical constants of Dhatryadi

KWaliTa		
Parameters	Result	
Loss on drying	15.19% w/w	
Ash value	8.48%	
Water-soluble extract	41.7% w/w	
Alcohol soluble extract	58.6% w/w	
рН	7	

Table 5: Chromatographic results of Dhatryadikwatha		
Conditions	Rf values (8 spots each)	
Short ultra violet (254 nm)	0.00, 0.08, 0.13, 0.54, 0.58, 0.68, 0.80, 0.89	
Long ultra violet (366 nm)	0.00, 0.08, 0.13, 0.51, 0.58, 0.72, 0.88, 0.94	



**Figure 2:** High performance thin layer chromatography evaluation of *Dhatryadi kwatha:* (a) Three-dimensional Graph: 254 nm and 366 nm of *Dhatryadi kwatha,* (b) chromatographic results (Peak display) of *Dhatryadi kwatha* at short ultra violet (254 nm), (c) chromatographic results (peak display)of *Dhatryadi kwatha* long ultra violet (366 nm)

for the identification of the medicinally important formulation of *Dhatryadi kwatha*. This work can be considered as the first step toward identifying the followed methods through HPTLC analysis. This is a preliminary analysis and meticulous nature along with the depiction is to be carried out.

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# REFERENCES

- 1. Shastri R, editor. Chakradatta 49/72. Varanasi: Chaukhamba Surbharati Prakashana; 2006. p. 196
- Tripathi YB, Singh VP, Sharma GM, Sinha RK, Singh D. X-ray diffraction and microscopic analysis of Tamra Bhasma: An Ayurvedic metallic preparation. Indian J Tradit Knowl 2003;2:107-17.
- Shailajan S, Menon S, Singh A. Quantitative analysis of piperine from Ayurvedic polyherbal formulations using reverse phase high performance liquid chromatography. Int J Pharm Bio sci 2009;1:1-10.
- Siddiqui A, Hakim MA. Format for the pharmacopoeial analytical standards of compound formulation. Workshop on Standardization of Unani Drugs, (appendix). New Delhi: Central Council for Research in Unani Medicine; 1995.

- Mukherjee PK. Quality Control of Herbal Drugs. 2<sup>nd</sup> Reprint ed. New Delhi: Business Horizons; 2007. p. 164-5.
- Anonymous. Indian Pharmacopeia. Appendix 8 (8.6). Vol. II. New Delhi: Government of India, Ministry of Health and Family Welfare, The Controller of Publication; 1996. p. A89.
- Anonymous. Indian Pharmacopeia. Appendix 8 (8.11). Vol. II. New Delhi: Government of India, Ministry of Health and Family Welfare, The Controller of Publication; 1996. p. A95.
- Anonymous. The Ayurvedic Pharmacopoeia of India, Part 1. Appendix 2 (2.2.3). 1<sup>st</sup> ed., Vol. VI. New Delhi: Government of India, Ministry of Health and Family Welfare; 2008. p. 242.
- Anonymous. The Ayurvedic Pharmacopoeia of India, Part 1. Appendix 2 (2.2.8). 1<sup>st</sup> ed., Vol. VI. New Delhi: Government of India: Ministry of Health and Family Welfare; 2008. p. 243.
- Anonymous. The Ayurvedic Pharmacopoeia of India, Part 1. Appendix 2 (2.2.7).1<sup>st</sup> ed., Vol. VI. New Delhi: Government of India: Ministry of Health and Family Welfare; 2008. p. 243.
- 11. Shukla VJ, Bhatt UB. Methods of Qualitative Testing of some Ayurvedic Formulations. Jamnagar: Gujarat Ayurvedic University; 2001.
- Anonymous. Ayurvedic Pharmacopoeia of India, Part-2. Appendices. 1<sup>st</sup> ed., Vol-2. New Delhi: Government of India, Ministry of Health of Family Welfare; 2008. p. 165-7.

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