EUROPEAN JOURNAL OF BIOMEDICAL AND PHARMACEUTICAL SCIENCES
http://www.ejbps.com

ISSN 2349-8870
Volume: 3
Issue: 10
433-437
Year: 2016

PHARMACOGNOSTICAL AND PHARMACEUTICAL EVALUATION OF *ASHWAGANDHADI VATI* - A FORMULATION FOR ASTHI KSHAYA AND ITS MANAGEMENT

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Article Received on 24/07/2016 **Article Revised on 14/08/2016** **Article Accepted on 04/09/2016**

ABSTRACT
Osteoporosis literally means porous bone. The medical community defines osteoporosis as a skeletal disorder in which bone strength is reduced as a result of loss of bone mass and through the deterioration of the bone architecture. The consequence of these changes is an increased fracture risk. No work has been carried out previously on *this Anubhuta formulation*. Hence an attempt has been made to study *Ashwagandhadi vati* by pharmacognostical, physico-chemical parameters and to develop HPTLC (High-Performance Thin Layer chromatography study) fingerprints of the compound formulation of *Ashwagandhadi vati.* Pharmacognostical work shows that presence of Lignified fibres through medullary rays of Ashwagandha, Border pitted vessel of Ashwagandha, Brown contant of Mandukaparni, Crystaline matirials of Shilajitu, Physico-chemical parameters shows that Ash value percentage4.8 % w/w, Loss on drying percentage2.61 % w/w and Water soluable extract percentage 0.246 % w/w. On performing HPTLC, the chromatogram of *Ashwagandhadi vati* showed 9 (nine) spotss at short waveUV 254nm; while at long wave UV 366 nm, the chromatogram showed 5 (Five) spots.

KEY WORDS: *Ashwagandhadi vati*, HPTLC, Pharmacognosy, Physicochemical analysis.

INTRODUCTION
Ayurveda is one of the most ancient medical sciences in the world. It describes the aspects of life, health, disease and its managements. Ayurveda names seven basic tissues (*Dhatu*), which are *Rasa, Rakta, Māmsa, Meda, Asthi, Majja* and *Shukra*. If any of these are not in balance it can cause various disorders.

Asthi Kshaya is the condition in which the *Asthi Dhatu* decreases. It occurs due to lack of *poshana* (nutrition) of the *Asthi Dhatu.* The function of *Asthi Dhatu* is to support the body and give shape to it. *Asthi Kshaya* vis-à-vis osteoporosis is a global problem that will increase in significance with the growing elderly population. There is no direct reference regarding the *Nidanas* of the *Asthi kshaya*, but since *Asthi dhatu* and *Vata Dosha* are inversely proportional to each other (*Aashrayaashrayee bhaava*) the *Vata Prakopaka Nidanas* can be taken as the *Nidanas* of *Asthi Kshaya*.

Osteoporosis literally means porous bone. The medical community defines osteoporosis as a skeletal disorder in which bone strength is reduced as a result of loss of bone mass and through the deterioration of the bone architecture. The consequence of these changes is an increased fracture risk.[1] When *Asthi Kshaya* occurs, *Asthi Vardhaka Dravyas* should be given.

The formulation *Ashwagandhadi vati* is formulated according to its contained drugs, which are well known as being effective in the treatment of osteoporosis. So it was selected as a treated group in this present study. No work has been carried out previously on *this Anubhuta formulation.*

Lack of standardization of poly-herbal formulations creates difficulty in validating the efficacy and maintaining quality of the product. Therefore, it is important to ensure the standard and quality right from the raw drugs to the finished product. Hence an attempt has been made to study *Ashwagandhadi vati* by pharmacognostical, preliminary phytochemical, physico-chemical parameters and to develop HPTLC (High-Performance Thin Layer chromatography study)

fingerprints of the compound formulation of *Ashwagandhadi vati*.

MATERIALS AND METHODS

Collection and authentication of raw drugs

All drugs were collected from the pharmacy of Gujarat Ayurved University, Jamnagar. The organoleptic and powder microscopy of drugs were carried out in Pharmacognosy Laboratory of I.P.G.T. & R.A. Drugs were confirmed to be authentic and of good-quality.

Pharmacognostical Study

Pharmacognostical analysis of *Ashwagandhadi vati* based on organoleptic characters and Microscopic studies, i.e, dissolving *Ashwagandhadi vati* in small quantity of distilled water, filtering through filter paper and the precipitate treated with or without stain to find out the characters and were later compared with the findings of individual ingredients of the *Ashwagandhadi vati*. The micro photographs were taken under Carl Zeiss Trinocular microscope attached with camera.[2,3]

Pharmaceutical Study: *Ashwagandhadi vati* was analyzed with appropriate protocols for standard physico-chemical parameters, such as aqueous soluble extract, alcohol soluble extract, pH, hardness, uniformity of weight, total ash, acid insoluble ash, loss on drying as per CCRAS recommendations at the Pharmaceutical Chemistry Laboratory, IPGT & RA.[4,5,6]

HPTLC: Methanol extract of *Ashwagandhadi vati* was spotted on pre coated silica gel GF 60_{254} aluminum plates by means of Camang Linomate V sample applicator fitted with a 100 µL Hamilton syringe. The mobile phase consisted of Chloroform: MeOH in a ratio of 9:1 v/v. After development densitometry scan was performed with a Camag T. L. C. scanner III in reflectance absorbance mode at 254nm and 366nm under control of Win CATS Software (V 1.2.1. Camag). Then the plate was sprayed with Vanillin Sulphuric acid followed by heating and then visualized in day light.[7]

OBSERVATIONS AND RESULTS

The initial purpose of the study was to confirm the authenticity of the drugs used in the preparation of *Ashwagandhadi vati*. For this, extract powder of all the four ingredients were subjected to organoleptic and microscopic evaluation separately to confirm the genuineness of all the drugs. Later, after the preparation of formulation, pharmacognostical evaluation was carried out.

Organoleptic evaluation: Organoleptic features like color, odour, taste and consistency of the *Ashwagandhadi vati* were recorded and are placed in **Table no 2**.

Microscopic evaluation: Microscopic evaluation was conducted by dissolving *Ashwagandhadi vati i*n the distilled water, then stained and studied under microscope for the following characters are observed Lignified fibres through medullary rays of Ashwagandha, Border pitted vessel of Ashwagandha, Brown contant of Mandukaparni, compound starch grains of Ashwagandha, Cork in surface view of Ashwagandha root, Crystaline matirials of Shilajitu, Epidermal cells of Mandukaparni, Epidermal cells with cluster crystals of Mandukaparni, Fragments of Fibres of Mandukaparni, Lifgnified fibres through medullary rays of Ashwagandha, Pitted vessels of Mandukaparni, Lignified Stone cell of Ashwagandha, simple starch grains with hilum, Stone cells of Ashwagandha and Trichome of Mandukaparni **Plate No.1 Fig A-N.**

Pharmaceutical study of *Ashwagandhadi vati*: Physico-chemical parameters like loss on drying, ash value, water and alcohol soluble extract etc were carried out and the results are depicted in **Table 3**.

High performance thin layer chromatography (HPTLC): On performing HPTLC, the chromatogram of *Ashwagandhadi vati* showed 9 (nine) peaks with maximum R_f values 0.4, 0.17, 0.35, 0.43, 0.59, 0.64, 0.68, 0. 91 and 0.97. at short wave UV 254nm; while at long wave UV 366 nm, the chromatogram showed 5 (Five) spots with maximum R_f values 0.05, 0.15, 0.34, 0.58 and 0.65 **Table 5 (Plate 2.Fig.1 - 2)**

Table – 1: Ingredients of *Ashwagandhadi vati*

Sl No:	Drug	Botanical Name	Part Used	Ratio
1	*Ashwagandha*	*Withania somnifera*	Root	2 parts
2	*Shilajatu*	*Black bitumen*		100mg
3	*Mandukparni*	*Centella asiatica(Linn)Urban*	*Panchanga*	1 part
4	*Dughdika*	*Euphorbia thymifolia*	Leaves, seeds	0.5 part

Table 2: Organoleptic characters of *Ashwagandhadi vati*

Characters	*Ashwagandhadi vati*
Colour	Light brown
Odour	Gritha
Taste	Bitter astrigent
Touch	Hard
weight	500mg

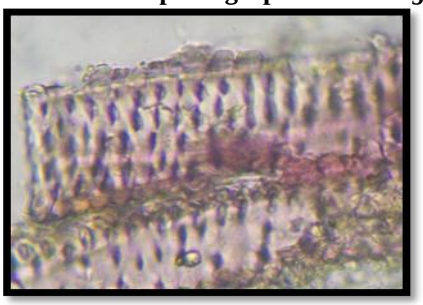
Table 3. Physico-chemical parameters of *Ashwagandhadi vati*

No.	Name of the Analysis	Value of. *Ashwagandhadi vati*
	Ash value percentage	4.8 % w/w
	Loss on drying percentage	2.61 % w/w
	Water soluable extract percentage	0.246 % w/w
	Alcohol soluble extract percentage	8.68% w/w
	Weight variation of Capsule	Average wt. 0.65gm Highest wt. 0.74gm Lowest wt. 0.4479gm
	pH value	4

Table 4: HPTLC results of *Ashwagandhadi vati*

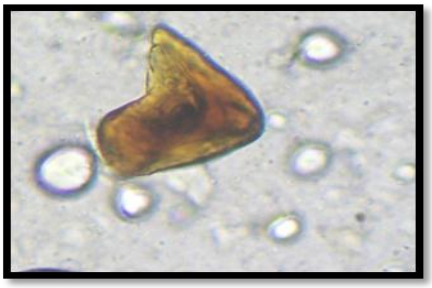
	254 nm		366nm	
HPTLC	**No. of Spots**	**Rf Value**	**No. of Spots**	**Rf Value**
	09	0.4, 0.17, 0.35, 0.43, 0.59, 0.64, 0.68, 0. 91, 0.97.	05	0.05, 0.15, 0.34, 0.58, 0.65

Plate 1. Microphotographs of *Ashwagandhadi vati*

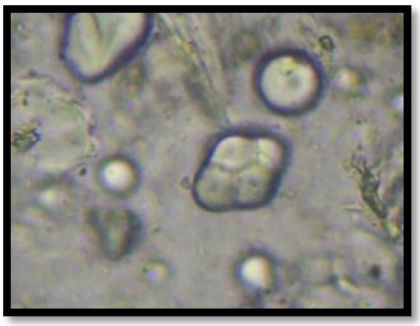


A. Lignified border pitted vessel of Ashwagandha-2

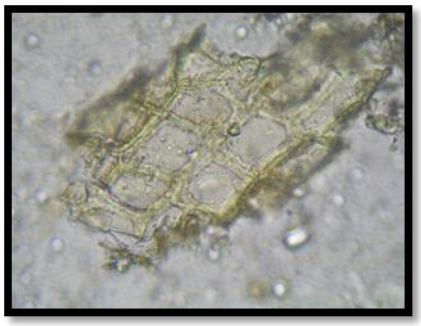
B. Border pitted vessel of Ashwagandha



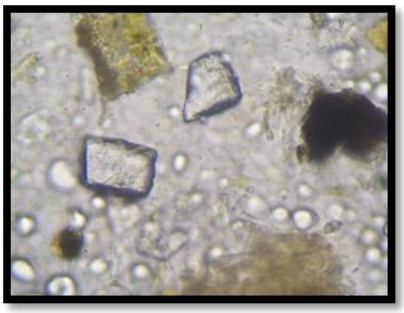
C. Brown contant of Mandukaparni



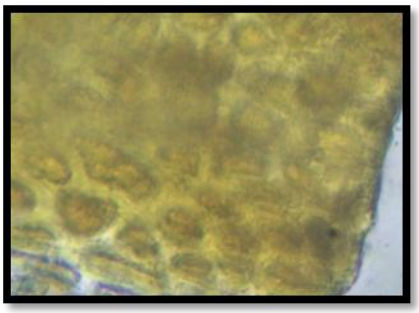
C. compound starch grains of Ashwagandha



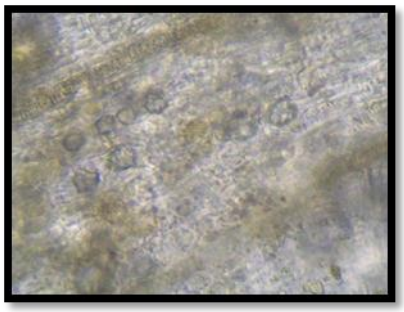
D. Cork in surface view of Ashwagandha root



E. Crystaline matirials of Shilajitu

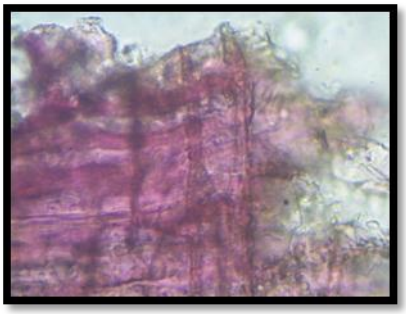


F. Epidermal cells of Mandukaparni



G. Epidermal cells with cluster crystals of Mandukaparni

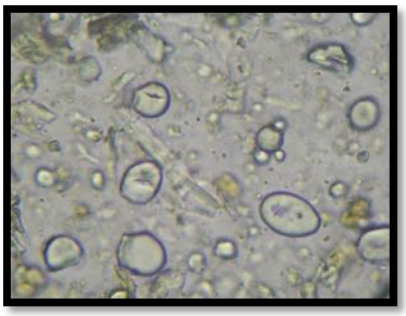
H. Fragments of Fibres of Mandukaparni



I. Lifgnified fibres through medullary rays of Ashwagandha

J. Lignified Stone cell of Ashwagandha

K. Pitted vessels of Mandukaparni

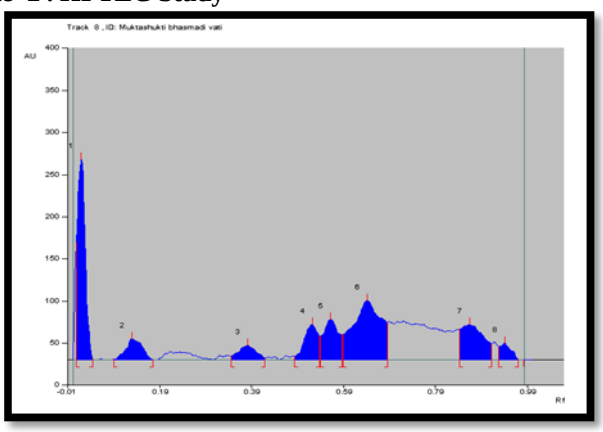


L. simple starch grains with hilum

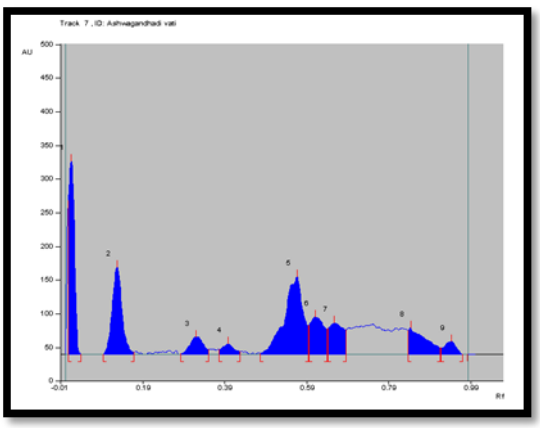
M. Stone cells of Ashwagandha

N. Trichome of Mandukaparni

Plate 2 . HPTLC Study



1. At 254nm



2. At 366nm

DISCUSSION

Ashwagandhadi vati, the formulated drug, found to be beneficial for hypertriglyceridemia has been analyzed. Study on *Ashwagandhadi vati* was a step towards pharmacognostical and pharmaceutical standardization of the drug. The presence of all contents of raw drugs in the final product showed the genuinity of the final product. All the pharmaceutical parameters analyzed has shown values permissible for the *Vati*. The physico-chemical parameters showed that percentage of water soluble extract was more than alcohol soluble extract. Ash value of the final product is 4.8 % w/w shows the presence of inorganic material. It also showed presence of slightly acidic nature of *Vati* which may help in augmenting the *Asthi kshaya (Osteoporosis)*. The phyto-chemical evaluation of *Ashwagandhadi vati* was done and it showed the presence of carbohydrates, steroids, glycosides, saponins, tannins and phenols. Thus it can be inferred that the drug may yield desired pharmacological action.

CONCLUSION

The ingredients were identified and authenticated pharmacognostically and were used for the preparation. The formulation was subjected to pharmacognostical study reveal genuineness as that all the ingredient microscopical characters were observed. Physico-chemical and HPTLC studies inferred that the formulation meets the minimum quality standards as reported in the API at a preliminary level. Though the ground work requisites for the standardization of *Ashwagandhadi vati* is covered in the current study, additional important analysis and investigations are required for the identification of all the active chemical

constituents of the test drug to substantiate the clinical efficacy. The inference from this study may be used as reference standard in further quality control researches.

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