



## Research Article

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### A PRELIMINARY PHYSICO-CHEMICAL ASSAY OF VAJIGANDHADI NIRUHA BASTI

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#### ABSTRACT

The Vajigandhadi Basti has been mentioned specifically for Gridhrasi (Sciatica) in Yogarathnakara Vatavyadi Chikitsa Adhikarana. Niruha is the one type of Basti (medicated enema) containing Kwatha (decoction of herbal drugs) as the chief constituent along with Madhu (honey), Saindhava (rock salt), Kalka (paste of herbs) and Sneha (oil). In Vajigandhadi Niruha Basti (VNB) Vajigandhadi Kwatha (decoction), Putoyavani Kalka (paste) and Vajigandhadi Taila (oil) are being used. The prepared VNB medicament subjected to physico-chemical and chromatographical parameters for standardization. Pharmacognostical identified features of coarse powder were systemically explained and physicochemical findings of prepared Basti such as specific Gravity, total solid contents, pH value were found to be 1.0390, 23.82% gm/ml, and 5.5 respectively. The chromatographic authentication of prepared VNB was done under pre-chromatographic derivatization technique. Microscopic characteristics like the border pitted vessels, stellate trichome, scleroid, oil globules, rhomboidal crystal, starch grains, prismatic crystals, annular vessel, black debris etc. showed the presence of mentioned ingredients in prepared VNB. The findings from this study will provide systemic evaluation and also encouraging towards the goal for standardizing VNB.

**Key words:** Chromatography, Gridhrasi, Pharmacognosy, Vajigandhadi Niruha Basti.

#### INTRODUCTION

Sciatica is a benign syndrome characterized especially by pain starting from the lumbar region and radiating down towards the lower limb. Ayurveda had identified this problem long back and named it 'Gridhrasi'. The word 'Gridhrasi' itself suggests the gait of the patient similar to Gridhra (vulture) due to pain<sup>1</sup>. Gridhrasi comes under 80 types of Nanatmaja Vatavyadhis<sup>2</sup>. Basti has been glorified as the definitive therapy to subdue the aggravated Vata and Vata Pradhan Vyadhis<sup>3</sup>. Acharya Charaka has mentioned Basti as a procedure in which the drugs that are being administered through rectal route, reach up to the Nabhi (Umbilical region), Kati (Lumber region), Parshva (Loin region) and Kukshi Pradesha (Abdomen), churn the accumulated Purisha and Dosha, spread the unctuousness (potency of the drugs) all over the body and easily come out along with churned Purisha and Doshas<sup>4</sup>. On the other hand standardization point of view to achieve quality of finished product entirely depends on the quality of the raw materials. Therefore, first step of standardization is the quality control aspects of raw material. It can be achieved by macroscopic and microscopic examination of the crude drugs. Preparing the VNB formulation with standard operating procedure which followed by Charak Samhita as SOS approach successfully. Another way the Vajigandhadi Taila has been mentioned in Yogarathnakara Vatavyadi Chikitsa Adhikarana, specifically indicated only for Gridhrasi and the form of administration has been told as Snehapana and Basti<sup>5</sup>. For

standardization point view this (VNB) have no valid document is not published. An unorthodox way a preliminary physico-chemical analysis of Gokshura Punarnava Basti has been published<sup>6</sup> successfully. Ayurvedic formulation standardization is focused more or less Pharmacopoeial monograph to validate their claim. This study has designed the valid road map for VNB in favour of physico-chemical (Density, Specific gravity, Total solid content and pH.) & high performance chromatographically profiling. In this way a sustainable identification tool for VNB formulation in context fingerprinting profile & physico-chemical constant.

#### AIMS AND OBJECTIVES

- To identify dried samples of ingredients powder of VNB macroscopically and microscopically.
- To preliminary analyze the prepared VNB by using different physico-chemical parameters.
- To develop the HPTLC fingerprinting profile.

#### MATERIALS AND METHODS

##### Collection of Raw Drug

All the raw drugs of VNB were obtained from Pharmacy, Gujarat Ayurved University, Jamnagar, India and all these were identified and authenticated in Pharmacognosy Laboratory, Institute for Postgraduate Teaching and Research in Ayurveda (IPGT & RA),

Gujarat Ayurved University, Jamnagar, India. Ingredients of VNB are depicted at [Table 1].

**Microscopical evaluation of powdered crude drugs**

Individual ingredient of the respective VNB formulation undertaken for powder-microscopic evaluation in standard operating procedure.

**Preparation method of Vajigandhadi Niruha Basti (VNB)**

The VNB was prepared by the classical method<sup>7</sup> in department of Panchakarma IPGT&RA, GAU, Jamnagar. Initially honey (50ml) and rock salt (5gm) were triturated in the Kharal, and then in respected order Vajigandhadi Taila (100ml) followed by Putoyavani Kalka (20gm) followed by Vajigandhadi Kwatha (350ml) were added and triturated till it emulsified properly.

Vajigandhadi Taila constituted Aswagandha, Bala, Bilva and Dasamoola (10 drugs as 1part) in equal proportion was prepared with Eranda Taila (Castor oil) base in RS and BK department, IPGT&RA, GAU, Jamnagar, India. The Vajigandhadi Kwatha was prepared with the same drugs i.e. Aswagandha, Bala, Bilva and Dasamoola. These drugs were taken in equal proportion to a total of 175gm then 8 times water (1400 ml) was being added to Dravya and this mixture was being heated until it became 1/4<sup>th</sup> (350 ml). The Putoyavani Kalka was prepared by taking Yavani,

Madanphala, Bilwa, Kushatha, Vacha, Shatpushpa, Musta and Pippali in equal proportion. In Vajigandhadi Basti, Kalka Dravya has not been mentioned so, Putoyavani Kalka was taken because as per Acharaya Vagbhatta, If Kalka Dravya has not been mentioned in Basti then Putoyavanyadi Kalka can be taken as universal Kalka<sup>8</sup>.

**Physico-chemical study**

The organoleptic and physico-chemical analysis of prepared VNB performed in authentic way according to standard protocol.

**HPTLC (High Performance Thin Layer Chromatography) evaluation**

Methanolic extract of VNB was spotted on pre-coated silica gel – GF 60<sub>254</sub> aluminium plate as 6 mm bands, 10 mm apart and 10 mm edge of the plates, by means of a CAMAG Linomat V sample applicator fitted with a 100µL Hamilton syringe. Toluene: Ethyl acetate: Acetic acid (7:2:1) 10ml combination was used as a mobile phase. The development distance was 7cm and development time was 30min. After development, Densitometry scanning was performed with a CAMAG TLC scanner III in reflectance absorbance mode at 254 nm and 366 nm under control of Win CATS software (V1.3.4 CAMAG).

**Table 1: Ingredients of Vajigandhadi Niruha Basti (VNB)**

Sr. No.	Drugs	Botanical name	Part used
1	Madhu (Honey)	<i>Mel depuratum</i>	----
2	Saindhav (Rock Salt)	Sodium chloride	----
<b>Vajigandhadi Kwatha and Taila</b>			
1	Vajigandha (Ashwagandha)	<i>Withania somnifera</i> Linn.	Root
2	Bala	<i>Sida cordifolia</i> Linn.	Whole plant
3	Bilva	<i>Aegle marmelos</i> Corr.	Bark
4	Agnimantha	<i>Premna mucronata</i> Roxb.	Stem
5	Shyonaka	<i>Oroxylum indicum</i> Vent	Bark
6	Patala	<i>Stereospermum suaveolens</i> DC	Bark
7	Gambhari	<i>Gmelina arborea</i> Linn.	Bark
8	Brihati	<i>Solanum indicum</i> Linn.	Whole plant
9	Kantakari	<i>Solanum surattense</i> Burm. f.	Whole plant
10	Shalaparni	<i>Desmodium gangeticum</i> DC.	Whole plant
11	Prishniparni	<i>Uraria picta</i> Desv.	Whole plant
12	Gokshura	<i>Tribulus terrestris</i> Linn.	Whole plant
<b>Putoyavani Kalka</b>			
1	Yavani	<i>Trachyspermum ammi</i> Linn.	Fruit
2	Madanphala	<i>Randia dumetorum</i> Poir.	Fruit
3	Bilwa	<i>Aegle marmelos</i> Corr.	Fruit
4	Kushatha	<i>Saussurea lappa</i> C.B. Clark	Root
5	Vacha	<i>Acorus calamus</i> Linn	Rhizome
6	Shatpushpa	<i>Anethum sowa</i> Roxb. ex Flem.	Fruit
7	Musta	<i>Cyperus rotundus</i> Linn.	Tubers
8	Pippali	<i>Piper longum</i> Linn.	Fruit

**Table 2: Identified Microscopic Characters of Ingredients of Vajigandhadi Kwatha and Taila**

Drugs	Identified Microscopic Characters	Drugs	Identified Microscopic Characters
Vajigandha (Ashwagandha)	Cork cells, Border Pitted Vessels, Starch Grains	Gambhari	Stone cells, Border pitted vessels
Bala	Stellate Trichome, Oil glouble, Romboidal Crystal,	Brihati	Stone cells, Pitted vessel with Starch Grains, Simple trichome
Bilva	Simple & compound starch, Fibre with crystal, Scleroid,	Kantakari	Stone cells, Multi branch Trichome with Fibers
Agnimantha	Cork cells, Rhomboidal crystal	Shalaparni	Trichome, Pitted & Annular vessel, Epidermal cells
Shyonaka	Pitted stone cells, Cigar shaped crystals	Prishniparni	Spiral vessels, Prismatic crystal & cork cells
Patala	Prismatic crystals, Crystal fibres	Gokshura	Prismatic crystals, Epidermal cells

**Table 3: Identified Microscopic Characters of Ingredients of Putoyavani Kalka**

Ingredients	Identified Microscopic Characters
Yavani	Brown content, Annular vessel, Endosperm
Madanphala	Group of Stone Cells, Epicarp Cells, Mesocarp cells
Bilwa	Simple & compound starch, Fiber with crystal, Pitted stone cells
Kushatha	Spiral vessel
Vacha	Scleriform vessel, Starch grains
Shatpushpa	Oil Globules
Musta	Lignified Fibre, Prismatic crystals, Oleoresin
Pippali	Brown content, Black debris

**Table 4: Organoleptic characters of Prepared Vajigandhadi Niruha Basti**

Parameter Studied	Observations
Colour	Dull Brown
Odour	Offensive
Consistency	Slightly thick Liquid

**Table 5: Physico-chemical parameters of Vajigandhadi Niruha Basti (VNB)**

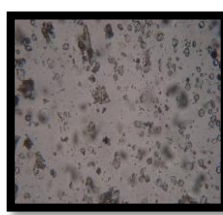
Parameter studied	Results DROP
Density	1.1874w/l gm/ml
Specific gravity	1.0390
Total solid contents	23.82%
pH (By pH strip)	5.5

**Table 6: R<sub>f</sub> values of Prepared Vajigandhadi Niruha Basti (VNB)**

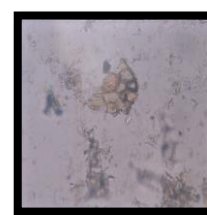
Sample	Visualize under short UV (254 nm)		Visualize under short UV (366 nm)	
	No. of spots	R <sub>f</sub> value	No. of spots	R <sub>f</sub> value
Vajigandhadi Niruha Basti	8	0.03, 0.16, 0.37, 0.55, 0.65, 0.76, 0.78, 0.95	8	0.02, 0.36, 0.37, 0.53, 0.56, 0.65, 0.76, 0.95



**Border Pitted Vessels (Ashwagandha)**



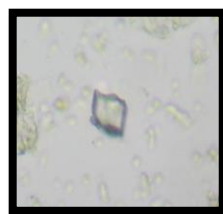
**Starch Grains (Ashwagandha)**



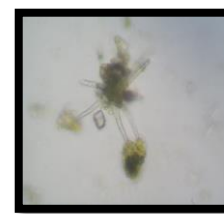
**Cork cells (Ashwagandha)**



**Oil globule (Bala)**



**Rhomboidal crystal (Bala)**



**Stellate trichome (Bala)**



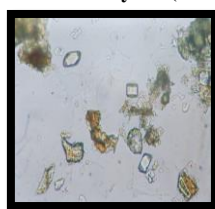
**Fibre with crystal (Bilwa)**



**Scleroid (Bilwa)**



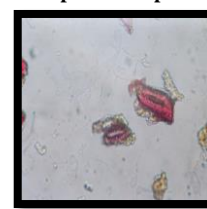
**Starch simple & compound (Bilwa)**



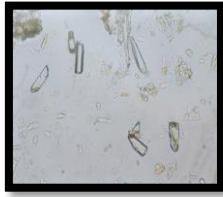
**Rhomboidal crystal (Agnimantha)**



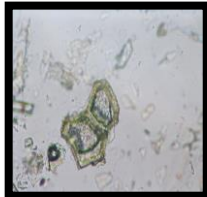
**Cork cells (Agnimantha)**



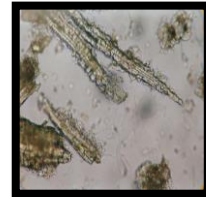
**Stone cells (Agnimantha)**



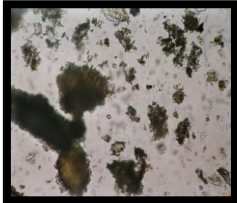
Cigar shaped crystal (Syonaka)



Pitted stone cell (Syonaka)



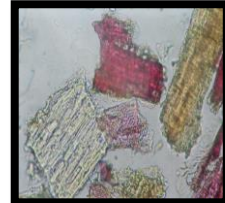
Crystal fibres (Patala)



Prismatic crystal (Patala)



Border pitted vessels (Gambhari)



Stone cells (Gambhari)



Pitted vessels with starch grains (Brihati)



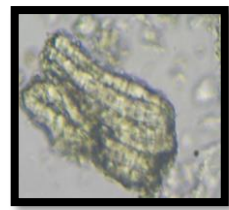
Stone cells (Brihati)



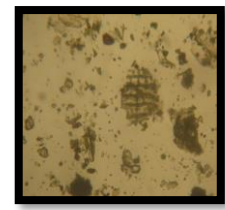
Simple trichome (Brihati)



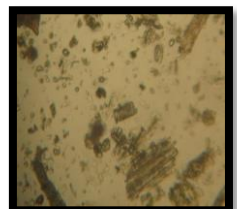
Multi branch trichome with fibres (Kantakari)



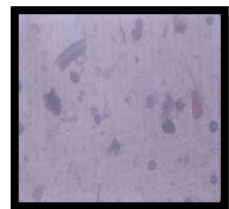
Stone cells (Kantakari)



Prismatic crystal & cork cells (Prishniparni)



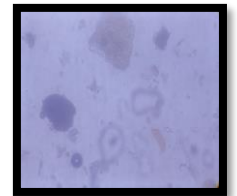
Spiral vessels (Prishniparni)



Pitted & Annular vessels (Shalaparni)



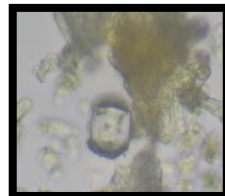
Trichome (Shalaparni)



Epidermal cells (Shalaparni)

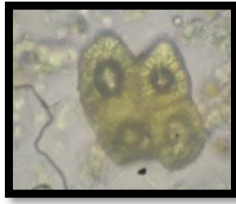


Epidermal cells (Gokshura)

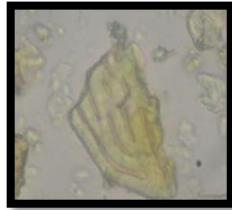


Prismatic crystals (Gokshura)

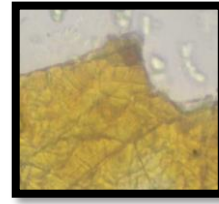
Plate 1 - Microphotographs of Vajigandhadi Kwatha and Taila Ingredients



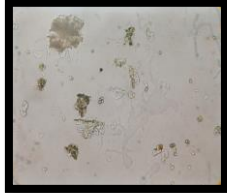
Group of Stone Cells  
(Madanaphala)



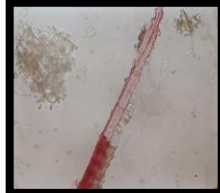
Epicarp cells (Madanaphala)



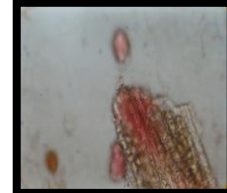
Mesocarp Cells (Madanaphala)



Simple & compound starch (Bilva)



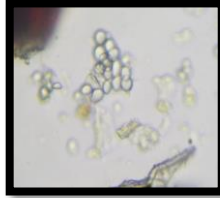
Fibre with crystal (Bilva)



Pitted stone cell (Bilva)



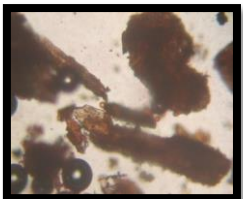
Spiral vessel (Kushtha)



Starch grains(Vacha)



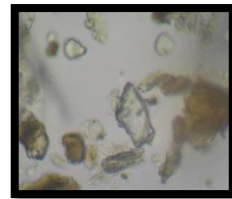
Scleriform vessel (Vacha)



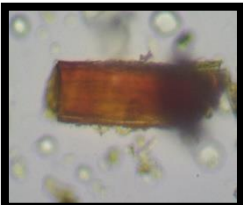
Oil Globules (Shatpushpa)



Lignified Fibre (Musta)



Prismatic crystals (Musta)



Oleoresin (Musta)



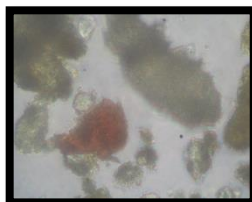
Brown content (Yavani)



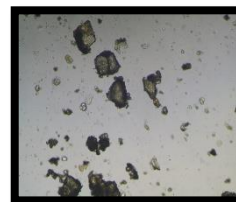
Annular vessel (Yavani)



Endosperm (Yavani)



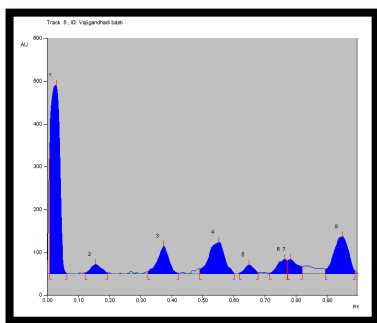
Brown content (Pippali)



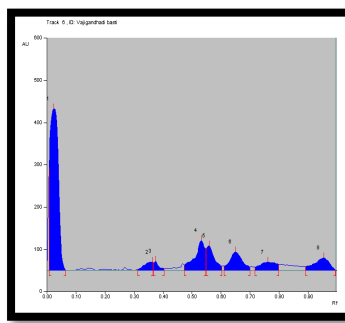
Black debris (Pippali)

Plate 2: Microphotographs of Putoyavani Kalka Ingredients



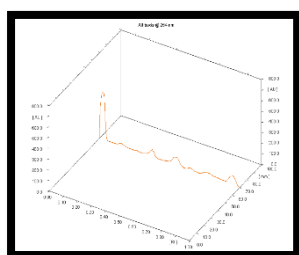


At 254nm

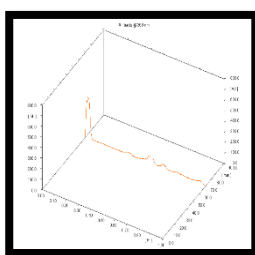


At 366nm

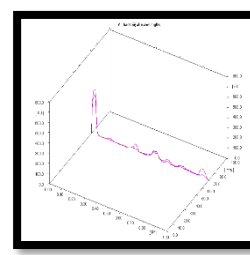
Plate 3: Densitogram of Prepared Vajigandhadi Niruha Basti (VNB) at 254 nm and 366nm



At 254nm



At 366nm



Comparison at 254nm & 366nm

Plate 4: Three dimensional (3D) Densitogram of prepared Vajigandhadi Niruha Basti (VNB) at 254nm, 366nm & Comparison at 254nm & 366nm

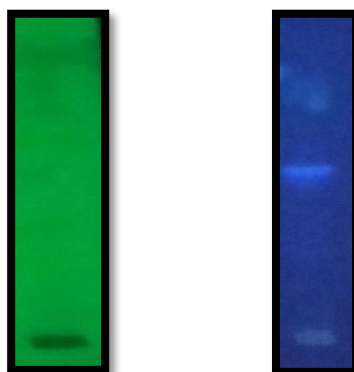


Plate 5: HPTLC finger prints of prepared Vajigandhadi Niruha Basti (VNB) at 254nm and 366nm (Fig. a, b)  
(a) At 254nm (b) At 366nm

## RESULTS AND DISCUSSION

### Microscopic Characters

Powder of the ingredients of Putoyavani Kalka, Vajigandhadi Taila and Kwatha were studied microscopically. The powder of the drug was dissolved with water. Then microscopy of the sample was done without stain and after staining with Phloroglucinol + HCl. The identified powder microscopic characters are placed at respective table [Table 2&3] and microphotographs are placed at respective plate [Plate 1&2]. All identified microscopic characteristics were equivalent to standard profile.

### Organoleptic characters of prepared VNB

Prepared VNB was dull brown in colour, slightly thick in consistency and had offensive odour [Table 4].

### Physico-chemical parameters

The prepared VNB was evaluated for various physico-chemical parameters like Density, Specific gravity, Total solid content and pH. The observed results are placed at Table 5. Density and specific gravity and pH is directly reflect the formulation stability condition. Total solid content revealed the dissolved materials in water (such as potassium, sodium etc.) moderate quantity to maintain the formulation status in favour of physico-chemical constant.

### HPTLC (High Performance Thin Layer Chromatography) evaluation

In this study VNB is well separated compact symmetrical bands in favour of chromophore sensitive component (Polyphenol, Flavonoid and quaternary alkaloids). This bio-ligand are responsible for formulation promising development condition.

HPTLC of VNB visualized under short UV (254nm) showed 8 spots at 0.03, 0.16, 0.37, 0.55, 0.65, 0.76, 0.78, 0.95 R<sub>f</sub> while under long UV (366nm) showed 8 spots at 0.02, 0.36, 0.37, 0.53, 0.56, 0.65, 0.76, 0.95 R<sub>f</sub> [Table 5/Plate 3 & 4]. Component represent by the R<sub>f</sub> 0.55, 0.65, 0.76 and 0.95 were common in both light exposure. The fingerprinting profile exemplified as an herb-print of this formulation.

## CONCLUSION

The present study was aimed to analyze the physico-chemical parameters of Basti form and also to develop HPTLC profile of VNB. The separation pattern of VNB is documented with help of prechromatographic derivative method in context of R<sub>f</sub> & densitogram. The Pharmacognostical evolution proved that ingredients were present in VNB. The VNB was found to be significant based on physico-chemical parameters. The findings from this study will provide systemic evaluation and shall also be encouraging towards the goal for standardization of VNB formulation. The study results may be used as the reference standard in further research undertakings of its kind.

## ACKNOWLEDGEMENT

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