

A Pharmaceutico - Analytical Study of *Nishothamadi Kashayam* in Dermatophytosis W.S.R to *DadruKushtha*

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Abstract: Introduction: Skin is the largest organ of human body. Incidence of fungal infections increases due to various reasons like unhygienic environmental conditions, poor sanitations, illiteracy, malnutrition, improper skin care, poverty etc. All the skin diseases in ayurveda have been classified under the broad heading of *kushta*. *Dadru* is one amongst them having *kapha* and *pitta* predominance. The main lakshanas of *dadru* include *kandu*, *raga*, *utsanna mandala* and *pidaka*. *Dadru* can be considered as dermatophytosis. Aim: Pharmaceutico analytical study of *Nishothamadikashaya* is against Dermatophytes causing *Dadru*. Materials and methods: *Nishothamadikashaya* made from *Haridra*, *Haritaki*, *Amalaki*, *Vibhitaki*, *Patola*, *Katuki*, *Vacha*, *Manjishta*, *Nimba* and sample of prepared medicine was analysed using following parameters as per the references available in protocol for testing published by Central Council for Research in Ayurvedic Studies. Results: All the analytical parameters of *Nishothamadi Kashaya* were within the normal limits and it has a substantial effect in reducing the symptoms caused by Dermatophytes. Conclusion: all of the constituents in *Nishothamadi Kashaya* shows more peaks in HPTLC studies indicates its potency. The ingredients are readily available, affordable and have no adverse effect. It demonstrates a fresh angle for using Ayurveda to treat Dermatophytosis.

Keywords: Dermatophytosis, *Nishothamadi Kashaya*, *Dadru*, Dermatophytes

1. Introduction

The knowledge of conversion of a raw material in to desirable medicine or dosage form is known as *Bhaishajya Kalpana*. It includes procurement of raw drug, processing of raw drug, preparation of medicine and its therapeutic application. Various other supporting factors like dose, route of administration, shelf life, palatability and modification according to the needs of patient's age, gender, *prakruthi* and season etc can be included under the umbrella of *Bhaishajya Kalpana*. The Pharmaceutical study includes mainly preparation of crude drugs and pharmaceutical processing in which drug ratio, liquid quantity, the intensity of the fire and its duration.

PanchavidhaKashaya Kalpanas are the basic pharmaceutical preparation and the most important form of *kalpanas*^[1]. In present clinical practice most abundantly used *Kalpana* among *PanchavidhaKashaya Kalpana* is *Kwatha Kalpana*^[2]. In this drug is boiled with 16 parts of water and reduced to 1/8th, then the filtrate is used as medicament. *Nishothamadikashayam* is one among the poly herbal formulation explained in the context of *kushtha*^[3]. Ingredients of this formulation are safe, easily available and economical and are also having *pitta-kapha hara*, *vranaropana* and *kushthaghna properties*.

Skin diseases are common manifestation in present era and more so frequent in the older age groups. *Dadru*, one of the most common but miserable variety of *Kushtha*, affects the population of all the age groups and stands as a challenge to different medical systems. Main symptoms of *Dadru* are *Kandu* (itching), *Daha* (burning sensation), *Rookshata* (dryness), *Raga* (erythema), *Pidaka* (eruptions), and *Utsanna mandala* (elevated circular skin lesions)^[4]. Dermatophytosis can be closely correlated with the

classical disease *Dadrukushtha*. Dermatophytosis is the most important cutaneous infection, generally confined to the layer of the skin and its appendages. It is also called as Ringworm or Tinea. The infection is called "ringworm" because it can cause an itchy, red, circular rash^[5]. In the present study, on taking into consideration all the above things, here an attempt was made to prepare *Nishothamadi Kashayato* ensure the process validation and to evaluate the formulation on the basis of Physico-chemical parameters. Analytical standards were generated by assessing the quality parameters of *kashaya*.

2. Materials and Methods

This includes two steps:

- Pharmaceutical study
- Analytical study

Pharmaceutical Study:

Pharmaceutical Study plays an important role to achieve successful treatment and it starts from the identification till the packing of the end products. Prior to use any herbs internally or externally for therapeutic intention, various procedure i.e., *Samskaras* are performed. Hence, *Kalpana* is the process through, which a substance is prepared into medicine form by using some raw materials according to the physician's requirement. In this part of study, detailed description regarding pharmaceutical process followed in the preparation of *Nishothamadikashaya*^[6] is discussed.

Aim: To prepare *Nishothamadikashaya* as per *Ashtanga hridayam*

Steps:

- Collection of Raw drugs.
- Processing of Raw drugs.

Volume 13 Issue 1, January 2024

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- Preparation of Nishothamadi Kashaya

Collection of raw drugs:

The raw drugs required for the preparation of medicine were collected from S.D.M Ayurveda pharmacy, Udupi and pharmaceutical study was conducted in *Rasashastra and Bhaishajyakalpana* practical Lab, S.D.M college of Ayurveda, Udupi

Processing of Drugs:

The raw drugs were checked for physical impurities and were subjected to washing and drying procedure and Haridra was washed and made in to small pieces

Equipments: Pounding machine, weighing machine, vessel, stirrer, cloth, gas stove, pyrometer

Table 1: Ingredients of *Nishothamadi Kashaya*

S. No	Ingredients	Parts used	Quantity
1	Haridra	Rhizome	10gms
2	Haritaki	Fruit	10gms
3	Amalaki	Fruit	10gms
4	Vibhitaki	Fruit	10gms
5	Patola	Root	10gms
6	Katuki	Rhizome	10gms
7	Vacha	Root	10gms
8	Manjishtha	Root	10gms
9	Nimba	Stem Bark	10gms

Method of preparation of *Nishothamadi Kashaya*

- The raw drugs were made into *yavakutachurna* using pounding machine.
- Haridra was made in to small pieces of length ½ inch by chopping.
- Then all these drugs were taken in a clean Stainless-steel Vessel having 5 litres capacity.
- Heat was given with LPG gas cylinder supplied gas stove, as fire was given and kept at minimum possible fire
- To these 1440ml of water was added.
- It was boiled till it reduced to 1/16th i.e. (180ml)
- Marking was done with measuring scale.
- The temperature of the *kashaya* during boiling was maintained in between 70° C- 90° C inside and outside 335° C- 350° C.
- It is then filtered using a thick cora white cloth.
- The filtered *kwatha* is taken in a separate vessel and the residue in the cloth is discarded.

Observations

- The colour of *Kwatha* changed from light brown to dark brown colour.
- The consistency of the liquid was gradually got increased.
- The smell of the ingredients could be appreciated; especially *Katuki* smell persisted while preparing *Kwatha*.
- The *yavakutachurna* obtained after the *paka* was found to be softer in nature
- The temperature noted on the day of preparation is 30-33° c and humidity 82%.
- Yield of *Nishothamadi Kashaya*: 180 ml

Table 2: Showing observations during the process of preparation of the sample

Parameters	
Kashaya Churna Qty.	84gms
Total qty. of water	1440 ml
Temperature given	70°c to 90°c
Time taken for the reduction	3 and half hrs
Total qty of <i>kashaya</i> obtained	180 ml

3. Analytical Study

Analytical study is the application of a process or a series of processes in order to identify and quantify a substance, the components of a solution or mixture, or the determination of the structures of chemical compounds and elements. Each and every drug substance has its own physical and chemical characteristics which help for separating it from other closely related drug. In present study an attempt was made to standardize the *Nishothamadi Kashaya* by employing various analytical parameters. sample of prepared medicine was analysed using following parameters as per the references available in protocol for testing published by Central Council for Research in Ayurvedic Studies (CCRAS)^[7]. Analytical study was carried out at in S.D.M. Centre for Research in Ayurveda and Allied sciences, Udupi.

Objectives:

- Assessment of organoleptic characteristics of *Nishothamadi Kashaya*.
- Physicochemical evaluation of *Nishothamadi Kashaya* on following parameters
 - pH
 - Refractive Index
 - Total solids
 - Specific gravity.
 - Foaming Index.

Assessment of Organoleptic Characters

Organoleptic evaluation of *Nishothamadikashaya* like colour, odour, taste, touch etc. conducted at pharmaceutical biochemistry laboratory of the institute.

Table 3: Organoleptic characteristics of *Nishothamadi Kashaya*

Sl. No	Parameters	Results
1	Colour / appearance	Dark brown
2	Sound	-
3	Odour	Characteristic smell of <i>katuki</i>
4	Taste	<i>Kashaya</i> rasa
5	Consistency	Liquid

Physico- chemical analysis

Physico-chemical analysis was carried out by following the parameters. Physico-chemical analysis i.e. Refractive Index^[8], Total solids^[9], Specific gravity^[10], Foaming Index and pH^[11] were mentioned in the table no. 3.

High Performance Thin Layer Chromatography^[12]

10ml of *Nishothamadikashaya* samples was partitioned with 20 ml butanol in a separating funnel and kept for 24hr. The butanol fraction was collected and filtered. The butanol was then made to evaporate on a waterbath and it is

dissolved in 10.0ml of methanol. 3 and 6µl of the above samples were applied on a pre-coated silica gel F254 on aluminum plates to a band width of 7 mm using Linomat 5 TLC applicator. The plate was developed Toluene: Ethyl acetate: Formic acid (5.0: 3.0: 0.5). The developed plates were visualized under short UV, long UV, under white light and derivatised with vanillin sulphuric acid spraying reagent, scanned under UV 254nm, 366nm, 540nm (white light) and 620nm (Following derivatisation). Rf, colour of the spots and densitometric scan were recorded.

4. Observation and Results

Organoleptic Evaluation

Various parameters of the material such as colour, odour, touch and taste etc. of the *nishottamadikashaya* were observed and recorded. Touch was analysed with the help of *Darshana, Sparshana, Ghrana* and *RasanaPareeksha* as mentioned in Ayurveda. Results were mentioned in the table no.3.

Physico-chemical Analysis

Physico-chemical analyses were carried out by following the parameters. Physico-chemical analysis i.e. Refractive Index, Total solids, Specific gravity, Foaming Index and pH were mentioned in the table no. 4.

HPTLC

High-performance thin-layer chromatography (HPTLC) is an invaluable quality assessment tool for the evaluation of botanical materials. The most critical parameter in achieving a separation with high selectivity is the choice of a stationary and mobile phase. In present work Toluene: ethyl acetate is used as mobile phase. Results of HPTLC are given in table no. 5,6 and densitogram are shown in graph no. 1,2&3

Table 4: Analytical results of *Nishothamadikashaya*:

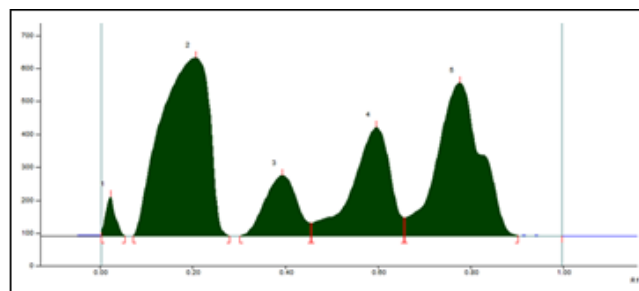
Parameter	Results n = 3% w/w <i>Nishothamadikashaya</i>
pH	6.0
Refractive index	1.36956
Specific gravity	1.0869
Total solids	26.17
Viscosity	5.48
Foaming index	28.12

HPTLC studies of *Nishothamadikashaya*

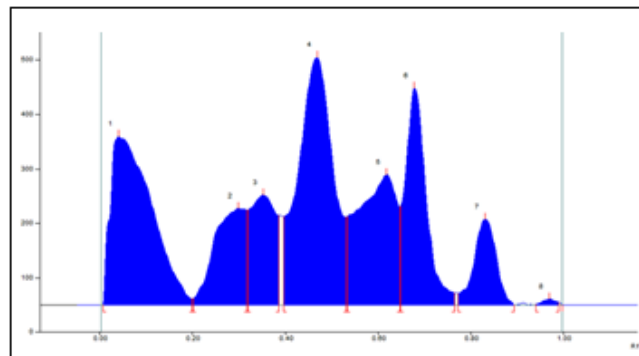
Table 5: Rf values of sample of Ethanol extract of *Nishothamadikashaya*

Short UV	Long UV	Post derivatisation
-	0.11 (F. blue)	-
0.17 (Green)	0.18 (F. blue)	-
-	0.27 (F. blue)	-
0.35 (Green)	0.33 (F. blue)	-
-	0.43 (F. blue)	0.42 (Pink)
0.55 (Green)	0.56 (F. blue)	-
-	0.61 (F. green)	0.62 (Purple)
0.69 (Green)	-	-
0.75 (Green)	0.74 (F. orange)	-

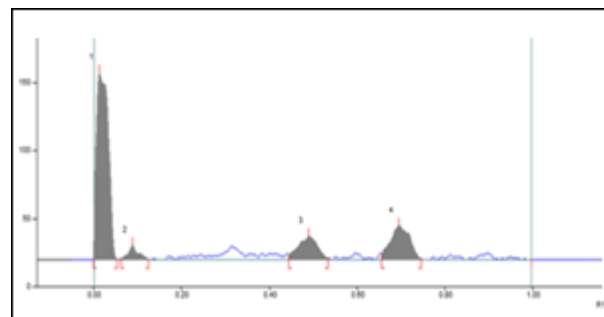
Densitometric scan of Ethanol fraction of *Nishothamadikashaya*



Graph 1: Overview Graph of *Nishothamadi Kashaya* at 254nm



Graph 2: Overview Graph of *Nishothamadi Kashaya* at 366nm



Graph 3: Overview Graph of *Nishothamadi Kashaya* at 620nm

Table 6: Showing Comparison of area and Peaks of *Nishothamadi Kashaya* at 254nm, 366nm and 620nm

Sample peak no.	<i>Nishothamadi Kashaya</i> at 254nm (area in AU)	<i>Nishothamadi Kashaya</i> at 366nm (area in AU)	<i>Nishothamadi Kashaya</i> at 620nm (area in AU)
1	1748.7	19962.0	2699.7
2	401514.5	8671.0	169.9
3	9153.3	7992.1	550.2
4	19206.5	24590.0	798.0
5	31608.0	14264.7	
6		12847.8	
7		5469.9	
8		219.1	

5. Discussion

Though Ayurveda is having its unique analytical approach towards drugs in present era, there is a necessity of

understanding a drug based on modern methodology of analysis also. So, there is a need to evaluate the drugs with various parameters.

Organoleptic characters - characters like colour, odour, touch, taste and appearance were observed using sense organs. *Nishothamadikashaya* was dark brown in colour with Astringent taste and characteristic odour.

Physio-chemical characters

pH value- Mean pH value of *Nishothamadikashaya* was shown slightly acidic in nature. It was facilitated the absorption of drug at the stomach level.

Refractive index- refractive index of a substance is defined as the ratio of the velocity of light in vacuum or air, compared to that in the substance. It indicates density of sample compared to air and liquid media. The refractive index of *nishothamadikashaya* is higher than the refractive index of universal Solvent. This indicates the presence of more solutes in it. The drugs added during the preparation of *kwatha* and its active principles might have been the reason for increased solutes.

Specific Gravity- Specific gravity of *Nishothamadikashaya* is slightly more than that of water. This indicates it has higher density than that of water. Density plays an important role in absorption of *kwatha*. Lesser the density of *kwatha* more will be the rate of penetration thus faster the rate of absorption.

Viscosity- It indicates the resistance of liquid to flow. Less viscous liquid has lesser resistance offered by the solutes, so it can be absorbed faster. The observed value of Viscosity of the sample was 5.48.

Total solids - Total suspended solids are the dry weight of suspended particles present in a known volume of sample. The observed value of total solids in *nishothamadikashaya* was 26.17.

Foaming index - it is indicating the amount of saponins present in the solution. solutions that have higher saponin content can permeabilize cell membranes without destroying them. The foaming index shown by *nishothamadi Kashaya* was 28.12.

HPTLC studies of *Nishothamadikashaya* -

In HPTLC, more peaks are found in *kashaya* at 366 nm. Under 254 nm we find 5 peaks and less number of peaks (4) are found at post derivatization phase (at 620 nm). Total area covered by the peaks is more in *kwatha* at 366nm. So, the number of peaks may indicate the effect in preserving the active ingredients of the drugs. It also indicates the potency of the sample. The difference in the number of peaks may point out the difference in the time and region of collection of drugs.

6. Conclusion

Phyto-chemical evaluation of *Nishothamadi Kashaya* showed the specific characters of ingredients which were used in the preparation. Organoleptic features, of *Nishothamadi Kashaya* were within the standard range. All

the Pharmaceutical parameters analysed within the allowable range and it may be used as standard reference for further research work and clinical studies

Acknowledgement

- 1) My deepest gratitude to guide Dr. Ravindra Angadi, Professor and Head, Department of Rasashastra and Bhaishajya Kalpana, SDMCA, Udupi for his guidance in completing this work and for his constant support
- 2) I express my deep sense of gratitude to co- guide Dr. Ashok Kumar B.N, Associate Professor, Department of Rasashastra and Bhaishajya Kalpana, SDMCA, Udupi for his constant support, encouragement and valuable guidance throughout this work
- 3) I solicit my gratitude to my co- guide Dr. Sushmitha V.S, Assistant Professor, Department of Rasashastra and Bhaishajya Kalpana, SDMCA, Udupi for her constant support, encouragement and guidance throughout this work
- 4) I am very grateful to Dr. Radhika Ranjan Geethesh.P, Associate Professor, Department of Rasashastra and Bhaishajya Kalpana, SDMCA, Udupi for his constant support, encouragement and guidance throughout this work

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