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Research Article

STANDARDIZATION OF *VARNYA DRAVYA* (COMPLEXION PROMOTERS) WITH SPECIAL REFERENCE TO *YASHTIMADHU* AND *MANJISHTA CHURNA* – AN ANALYTICAL STUDY

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KEYWORDS: *Varnya Dravya*, Complexion Promoters, *Yashtimadhu*, *Glycirrhizia glabra*, *Manjishtha*, *Rubia Cordifolia.*

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ABSTRACT

Standardization of drugs refers to the confirmation of its identity and determination of the quality and purity. Herbal drug technology is used for converting botanical materials into medicines, where standardization and quality control with proper integration of modern scientific techniques and traditional knowledge are important. However, the quality control and quality assurance still remains a challenge because of the high variability of chemical components involved. Most of the pharmaceutical industries are using substitute drugs in place of authentic drugs. So to manufacture and deliver the best quality drugs, it is essential to authenticate the raw drugs. Keeping the current inclination in mind, *Varnya dravya* or the complexion promoting drugs such as *Yashtimadhu* (*Glycirrhizia glabra*) and *Manjishtha* (*Rubia Cordifolia*) *churna* were subjected for standardization procedures. From the current study, genuinity indicating parameters for both *Yashtimadhu churna* (powder) and *Manjishtha churna* were derived.

INTRODUCTION

Beauty, specially fairness of skin, is a subject of socio-medical importance and has given rise to many skin-lightening procedures such as dermabrasion, ultrasound, and laser therapy.[1] The unique, effective and long lasting concept of beauty in Ayurveda has led to the emergence of Ayur-cosmaceuticals. The concepts of *Varṇa, Chāyā, Prabhā* dealt in Ayurveda are innate entities of beauty. The word *Varṇa* in Sanskrit means "outward appearance, exterior form, figure, shape, colour", "colour of the face", "good colour or complexion, lustre, beauty.[2] *Varṇa* is not just colour but it includes all the parameters of healthy and radiant skin.[3]The term *Varnya* refers to that which imparts Varna[4] (skin colour) i.e., it acts as an instrument to restore and retain the natural hue, texture and tone of the skin. These *Varnya dravya* (complexion promoters) are not to convert the inherent colour and complexion into fairer one, but to exemplify the abnormal colour which is changed by some disturbance in normal state. Ayurvedic cosmetics are in use and practice since thousands of years in India, without any side effects and are well proven and documented. The analysis of many herbal ingredients using modern scientific technologies has led to the identification of phytochemical components in Indian herbs, which deliver functional benefits anti dandruff, deodorant, age-defying properties etc.[5]

In recent years, more people throughout world are turning to use medicinal plant products in healthcare system.[6] Ancient Indian literature comprises a remarkably broad definition of medicinal plants and considers all plant parts to be potential sources of medicinal substances.[7] However, a key obstacle which has hindered the acceptance of these traditional medicines in the developed countries is the lack of documentation and rigorous quality control. There is a need for documentation of research work carried out on traditional medicines. With this backdrop, it becomes extremely important to make an effort

towards standardization of the plant-based medicines.[8]

Standardization of drug means confirmation of its identity, quality and purity throughout all phases of its cycle i.e., shelf life, storage, distribution and use by various parameters.[9] Standardization of herbal medicines is the process of prescribing a set of standards or inherent characteristics, constant parameters and definitive qualitative and quantitative values that carry an assurance of quality, efficacy, safety, and reproducibility. Quality of raw materials, good agricultural practices, and good manufacturing practices play fundamental roles in guaranteeing the quality and stability of herbal preparations.[10] World Health Organization (WHO) has published guidelines to ensure the reliability and repeatability of research on herbal medicines.[11]

Most of the pharmaceutical industries are using substitute drugs in place of authentic drugs. So, to manufacture and deliver the best quality drugs, it is essential to authenticate the raw drugs. Keeping the current inclination in mind, *Varnya dravyas* or the complexion promoting drugs such as *Yashtimadhu* (*Glycirrhizia glabra*) and *Manjishtha* (*Rubia Cordifolia*) *churna* were subjected for standardization procedures. From the current study, genuinity indicating parameters for both *Yashtimadhu churna* (powder) and *Manjishtha churna* were derived.

MATERIALS AND METHODS

Phytochemical tests like tests for alkaloids, steroids, saponins, tannins, flavonoids, phenol, coumarins, triterpenoids, carboxylic acid, resin, quinine and HPTLC were carried out as per the WHO guidelines, Ayurvedic Pharmacopoeia and Indian Pharmacopoeia.

Materials

Yashtimadhu (*Glycirrhizia glabra*) powder and *Manjishtha* (*Rubia Cordifolia*) powder were collected from SDM pharmacy, Udupi, Karnataka state, India.

Design

The studies were done at SDM Centre for Research in and Allied Sciences, Kuthpady, Udupi, Karnataka state, India as per standard procedure.

Methodology

1. Powder microscopy

A pinch of the sample was mounted on a microscopic slide with a drop of glycerin-water. Characters were observed using Zeiss AXIO trinocular microscope attached with Zeiss AxioCam camera under bright field light. Magnifications of the figures are indicated by the pre-calibrated scale-bars using Zeiss Axio Vision software.

2. Loss on drying at 105°C

10 g of sample was placed in tared evaporating dish. It was dried at 105˚C for 5 hours in hot air oven and weighed. The drying was continued until difference between two successive weights was not more than 0.01 after cooling in desiccator. Percentage of moisture was calculated with reference to weight of the sample.

3. Total Ash

2 g of sample was incinerated in a tared platinum crucible at temperature not exceeding 450˚C until carbon free ash is obtained. Percentage of ash was calculated with reference to weight of the sample.

4. Acid insoluble Ash:

To the crucible containing total ash, add 25ml of dilute HCl and boil. Collect the insoluble matter on ashless filter paper (Whatmann 41) and wash with hot water until the filtrate is neutral. Transfer the filter paper containing the insoluble matter to the original crucible, dry on a hot plate and ignite to constant weight. Allow the residue to cool in suitable desiccator for 30 mins and weigh without delay. Calculate the content of acid insoluble ash with reference to the air dried drug.

5. Water soluble ash

Boil the ash for 5 min with 25 ml of water; collect insoluble matter on an ashless filter paper, wash with hot water, and ignite for 15 min at a temperature not exceeding 450°C. Subtract the weight of the insoluble matter from the weight of the ash; the difference in weight represents the water soluble ash with reference to the air-dried sample.

6. Alcohol soluble extractive

Weigh accurately 4 g of the sample in a glass stoppered flask. Add 100 ml of distilled Alcohol (approximately 95%). Shake occasionally for 6 hours. Allow to stand for 18 hours. Filter rapidly taking care not to lose any solvent. Pipette out 25ml of the filtrate in a pre-weighed 100 ml beaker. Evaporate to dryness on a water bath. Keep it in an air oven at 105°C for 6 hours, cool in desiccator for 30 minutes and weigh. Calculate the percentage of Alcohol extractable matter of the sample. Repeat the experiment twice, and take the average value.

7. Water soluble extractive:

Weigh accurately 4 g of the sample in a glass stoppered flask. Add 100 ml of distilled water, shake occasionally for 6 hours. Allow to stand for 18 hours. Filter rapidly taking care not to lose any solvent. Pipette out 25ml of the filtrate in a pre-

weighed 100 ml beaker. Evaporate to dryness on a water bath. Keep it in an air oven at 105°C for 6 hours. Cool in a desiccator and weigh. Repeat the experiment twice. Take the average value.

8. HPTLC:

1g of Choorna (powder) was extracted with 10 ml of *alcohol*. 5 and 10µl of the above extract was 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 in Toluene: Ethyl acetate(8:2). The developed plates were visualized in UV 254, 366, and then derivatised with vanillin sulphuric acid and scanned under UV 254 and 366 nm. R_f, colour of the spots and densitometric scan were recorded.

9. Preliminary phytochemical tests

Tests for alkaloids

a. ***Dragendroff's test:*** To a few mg of extract dissolved in alcohol, a few drops of acetic acid and Dragendroff's reagent were added and shaken well. An orange red precipitate formed indicates the presence of alkaloids.

b. ***Wagners's test:*** To a few mg of extract dissolved in acetic acid, a few drops of Wagner's reagent was added. A reddish brown precipitate formed indicates the presence of alkaloids.

c. ***Mayer's test:*** To a few mg of extract dissolved in acetic acid, a few drops of Mayer's reagent was added. A dull white precipitate formed indicates the presence of alkaloids.

d. ***Hager's test:*** To a few mg of extract dissolved in acetic acid, 3 ml of Hager's reagent was added, the formation of yellow precipitate indicates the presence of alkaloids.

Tests for carbohydrates

a. ***Molisch's test:*** To the extract, 1 ml of *α*-naphthol solution and conc. sulphuric acid were added along the sides of test tube. Violet colour formed at the junction of the two liquids indicates the presence of carbohydrates.

b. ***Fehling's test:*** A few mg of extract was mixed with equal quantities of Fehling's solution A and B. The mixture was warmed on a water bath. The formation of a brick red precipitate indicates the presence of carbohydrates.

c. ***Benedict's test:*** To 5 ml of Benedict's reagent, a few mg of extract was added, and boiled for two minutes and cooled. Formation of a red precipitate indicates the presence of carbohydrates.

Test for steroids

a. ***Libermann-Burchard test:*** To the extract was dissolved in chloroform, 1 ml of acetic acid and 1 ml of acetic anhydride were added, then heated on a water bath and cooled. Few drops of conc. Sulphuric acid were added along the sides of the test tube. Appearance of bluish green colour indicates the presence of steroids.

b. ***Salkowski test:*** The extract was dissolved in chloroform and equal volume of conc. Sulphuric acid was added. Formation of bluish red to cherry red colour in chloroform layer and green fluorescence in the acid layer indicates the presence of steroids.

Test for saponins

To a few mg of extract, distilled water was added and shaken. Stable froth formation indicates the presence of saponin.

Test for tannins

To the extract, a few drops of dilute solution of ferric chloride was added, formation of dark blue colour shows the presence of tannins.

Test for flavonoids

Shinoda's test: To the extract in alcohol, a few magnesium turnings and few drops of conc. hydrochloric acid were added and heated on a water bath. Formation of red to pink colour indicates the presence of flavonoids.

Test for phenol

To the extract in alcohol, added two drops of alcoholic ferric chloride. Formation of blue to blue black indicates the presence of phenol.

Test for coumarins

To the extract in alcohol, a few drops of 2 N sodium hydroxide solution was added. Dark yellow colour formation indicates the presence of coumarins.

Test for triterpenoids

The extract was warmed with tin bits and few drops of thionyl chloride. Formation of pink colour indicates the presence of triterpenoids.

Test for carboxylic acid

Extract dissolved in water is treated with sodium bicarbonate. Brisk effervescence indicates the presence of carboxylic acid.

Test for resin

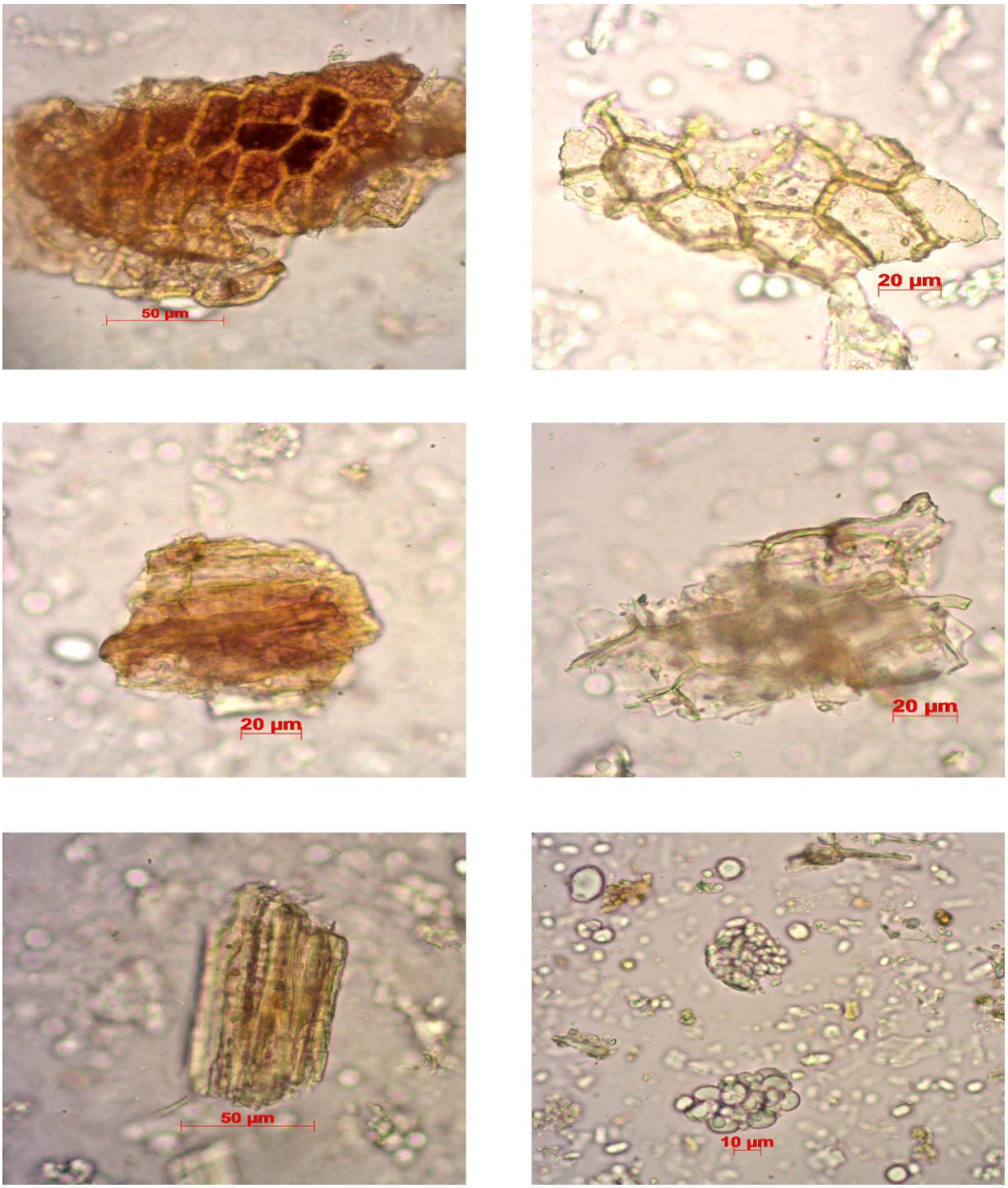
Few mg of the sample was mixed with water and acetone. Turbidity indicates the presence of resin.

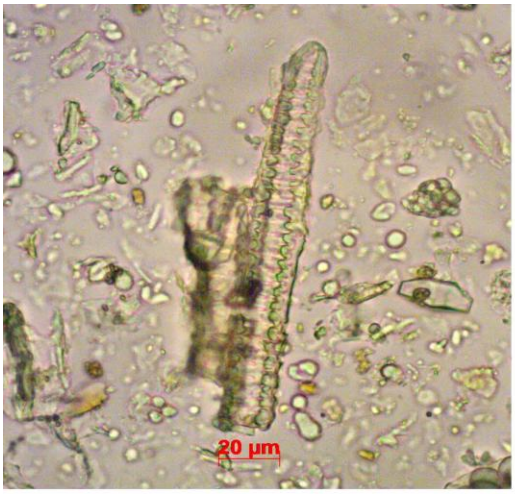
Test for quinine

A few mg of alcohol extract was treated with 0.5% of sodium hydroxide. Deep coloration like pink, purple or red indicates the presence of quinine.

RESULTS

Figure 1: Powder microscopy of *Yashtimadhu churna*

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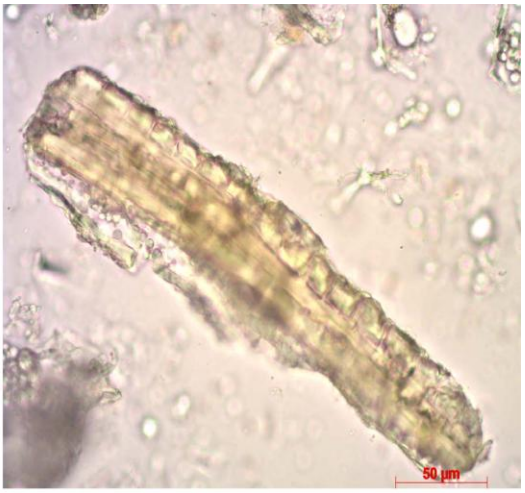
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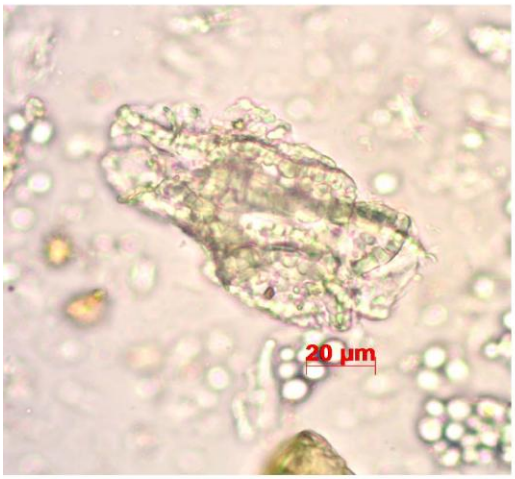
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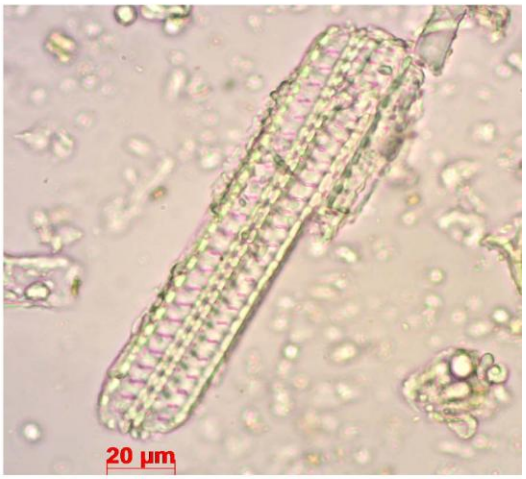
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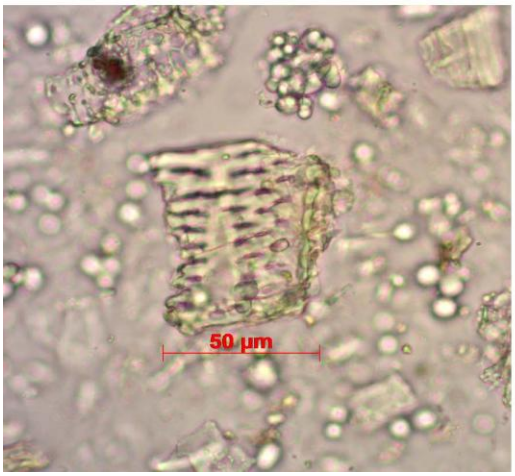
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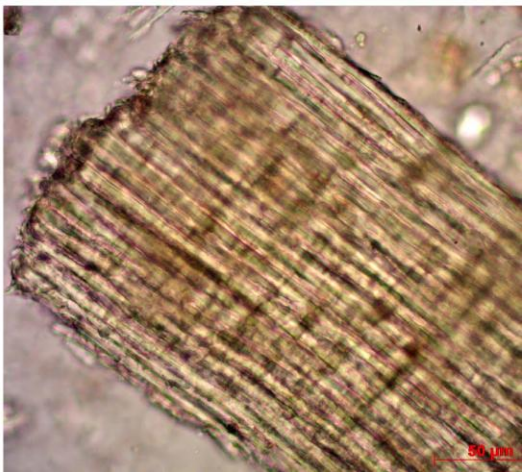
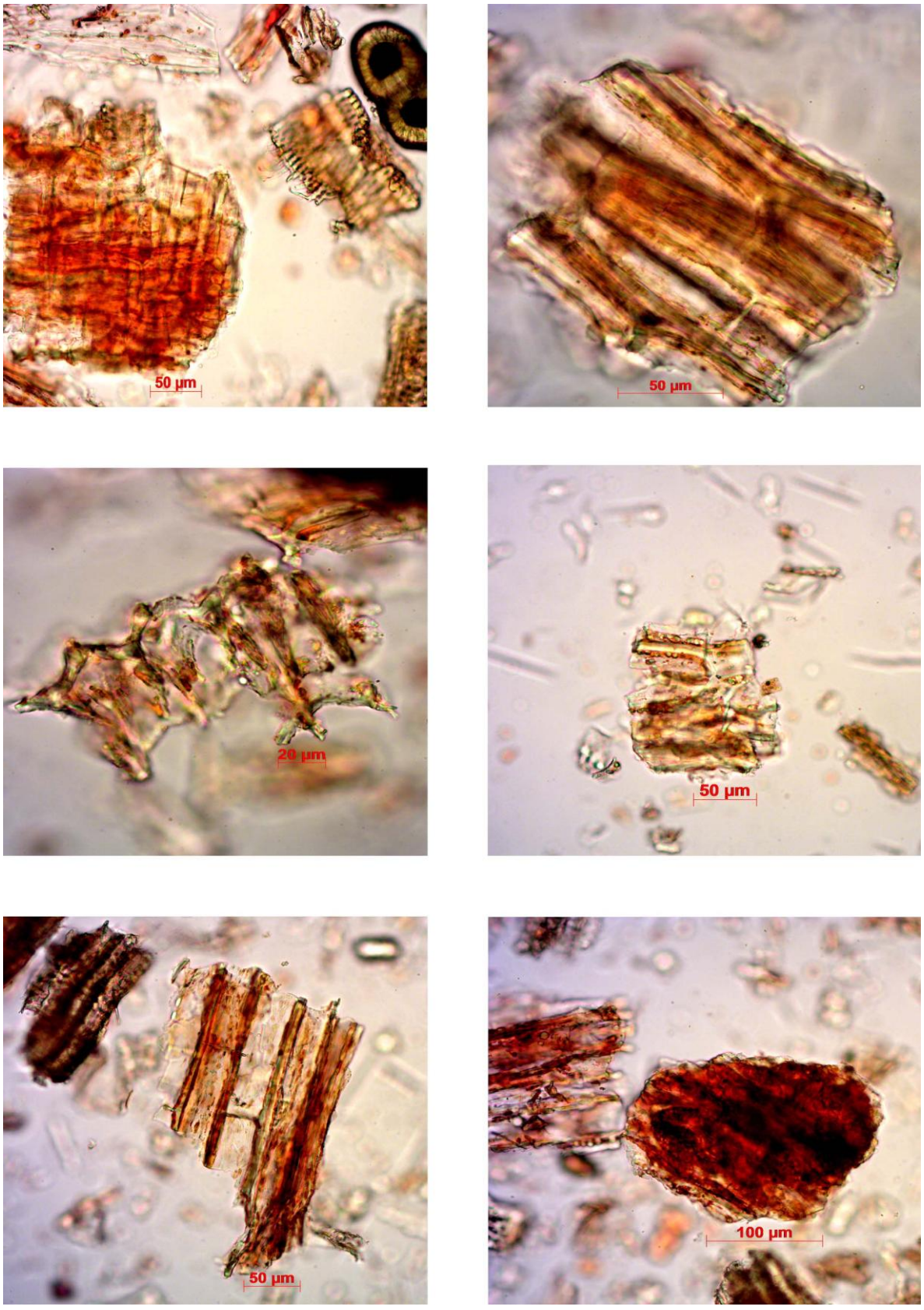
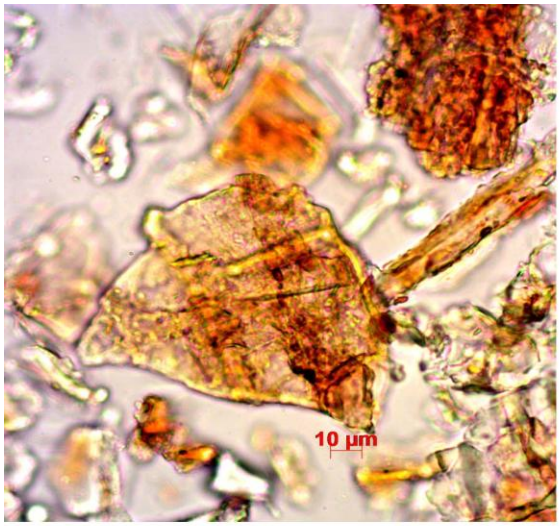


Figure 2: Powder microscopy of *Manjishta churna*

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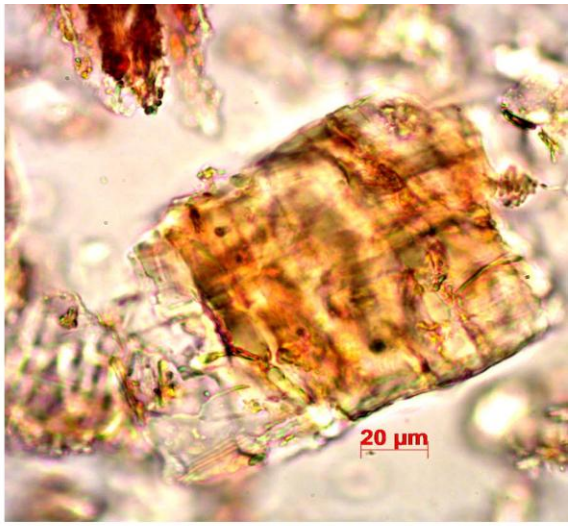

20 µm


20 µm


20 µm


50 µm


20 µm

Table 1: Results of preliminary phytochemical tests

Tests	**Colour if positive**	***Yashtimadhu churna***	***Manjishta churna***
Alkaloids			
Dragendrof's test	Orange precipitate	Orange Red Solution	Orange Red Solution
Wagners test	Red precipitate	Reddish Brown Colour	Reddish Brown Colour
Mayers test	Dull white precipitate	Light Yellow Colour	Light Yellow Colour
Hagers test	Yellow precipitate	Yellow Colour	Light Yellow Colour
Steroids			
Liebermann-buchard test	Bluish green	Green colour	Green Colour
Salkowski test	Bluish red to cherry red	Reddish Brown at junction	Reddish Brown at junction
Carbohydrate			
Molish test	Violet ring	Violet ring	Violet ring
Fehlings test	Brick red precipitate	Blue colour	Brick red precipitate
Benedicts test	Red precipitate	Green colour	Red precipitate
Tannin			
With $FeCl_3$	Dark blue or green or brown	Brown colour solution	Brown colour solution
Flavanoids			
Shinoda's test	Red to pink	Reddish Brown colour	Yellow Colour
Saponins			
With $NaHCO_3$	Stable froth	Little Froth	No Froth formed
Triterpenoids			
Tin and thionyl chloride test	Pink	Brown precipitate	Yellow Colour
Coumarins			
With 2 N NaOH	Yellow	Dark Red Colour	Dark Red Colour
Phenols			
With alcoholic ferric chloride	Blue to blue black, brown	Brown colour solution	Brown colour solution
Carboxylic acid			
With water and $NaHCO_3$	Brisk effervescence	No brisk effervescence	No brisk effervescence
Resin			
With aqueous acetone	Turbidity	Little turbidity	No Turbidity
Quinone			
5% NaOH	Pink/purple/red	Dark Red Colour	Red Colour

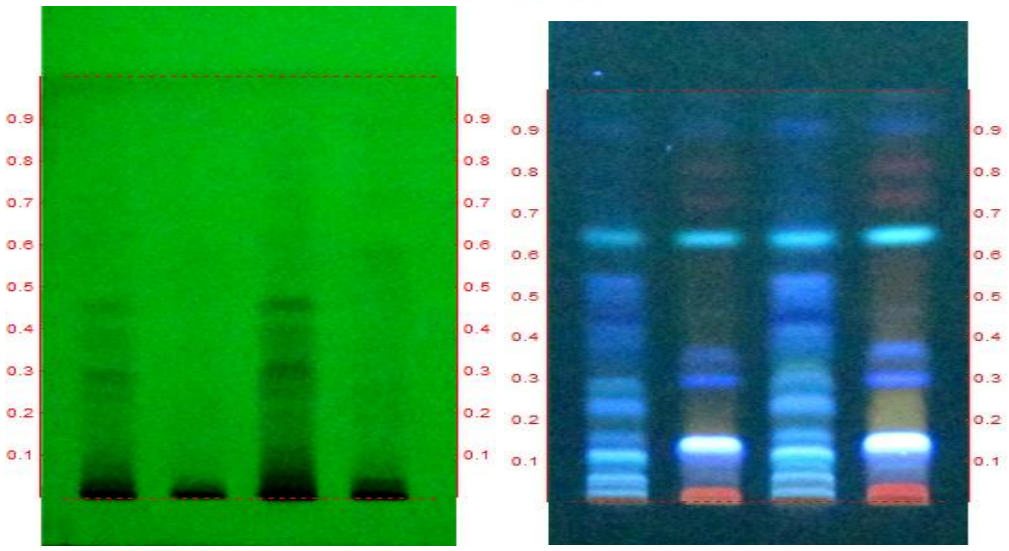
Table 2: Summary of preliminary phytochemical tests

Test	***Yashtimadhu churna***	***Manjishta churna***
Alkaloid	–	–
Carbohydrate	+	+
Carboxylic acid	–	–
Coumarins	–	–
Flavanoids	+	–
Phenol	+	+
Quinone	+	+
Resins	+	–
Steroid	+	+
Saponins	+	–
Tannin	+	+
Terpenoid	–	–

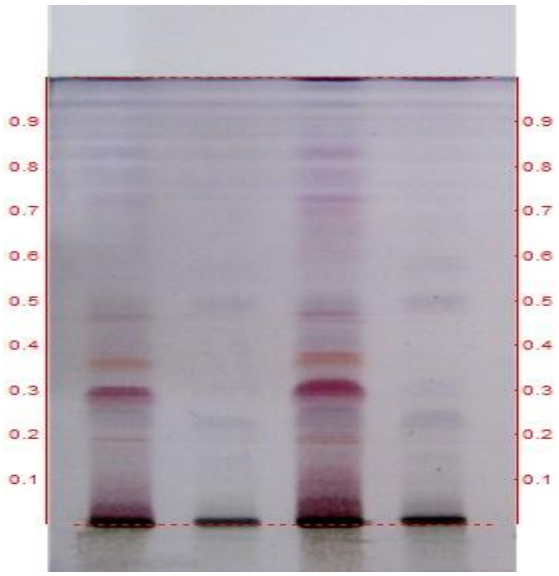
Table 3: Results of standardization parameters

Parameter	**Results n = 3 %w/w**	
	Yashtimadhu churna	***Manjishta churna***
Loss on drying At 105˚C	8.668	11.426
Total Ash	7.932	6.222
Acid Insoluble Ash	1.388	0.494
Alcohol soluble extractive	7.031	2.93
Water soluble extractive	19.009	10.45

Figure 3: HPTLC photo documentation of ethanol extract of *Yasthimadhu Churna & Manjishtachurna*



At 254 nm **At 366 nm**



After post derivatization

Track 1- *Yashtimadhu churna* – 4 µl
Track 2- *Manjishta churna* – 4 µl
Track 3- *Yashtimadhu churna* – 8 µl
Track 4- *Manjishta churna* – 8 µl

Solvent system: Toluene: Ethyl Acetate (8:2)

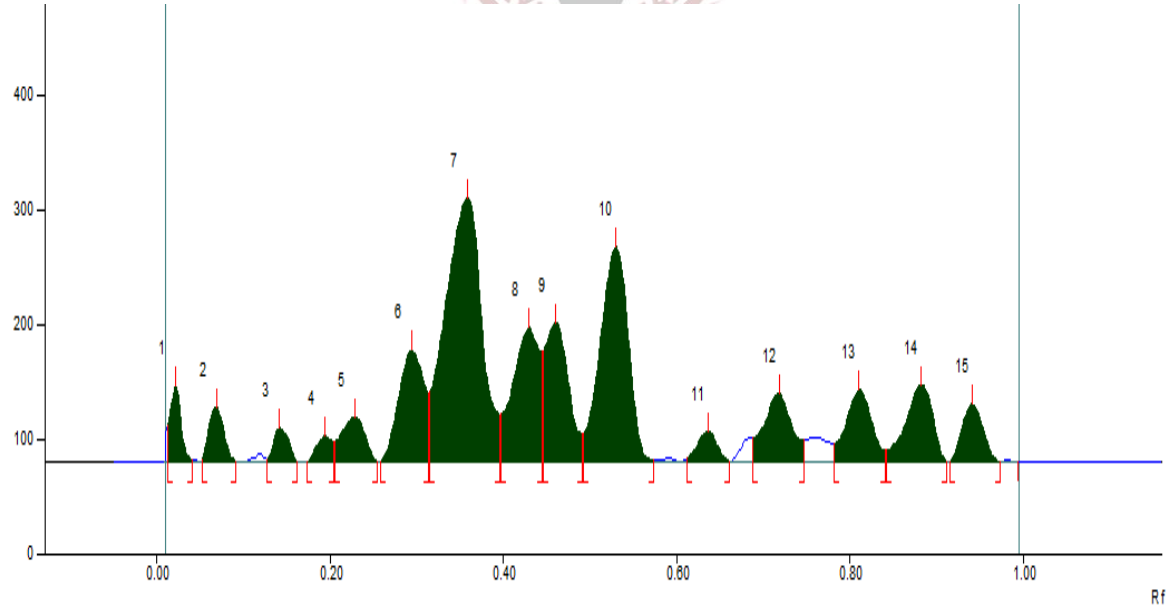
Table 4: R_f values of *Yashtimadhu churna & Manjishta churna*

At 254 nm		**At 366 nm**		**After post derivatisation**	
Yashtimadhu churna	***Manjishta churna***	***Yashtimadhu churna***	***Manjishta churna***	***Yashtimadhu churna***	***Manjishta churna***
-	-	0.03(F Blue)	0.03(F Red)	-	-
0.05 (D Grreen)	0.05 (D Green)	-	-	0.05 (Red)	0.05(L Violet)
-	-	0.08(F Blue)	0.08(F Blue)	-	-
-	-	0.09(F Blue)	-	-	-
0.11 (Green)	0.11(L Green)	-	-	-	0.11(L Violet)
-	-	0.13(F Blue)	-	0.13(L Brown)	-
-	-	-	0.16 (F Blue)	-	0.16(L Violet)
-	-	0.18(F Blue)	-	-	-
0.20 (L Green)	0.20(L Green)	-	-	0.20(Brown)	-
-	-	-	0.22(F Brown)	-	-
-	-	-	-	-	0.25(L Violet)
0.26(L Green)	0.26(L Green)	0.26(F Blue)	-	0.26(L Violet)	-
-	-	-	0.27(F Brown)	-	-
-	-	0.31(F Blue)	0.31(F Violet)	-	-
0.32(Green)	-	-	-	-	0.32(L Violet)
-	-	0.33(F L Green)	-	0.33(Red)	-

-	-	0.38(F Violet)	0.38(F Violet)	0.38(Brown)	-
0.41(L Green)	-	-	-	-	-
-	-	0.43(F Violet)	-	0.43(L Violet)	-
0.47(Green)	-	0.47(F Violet)	0.47(F Brown)	0.47(L Pink)	-
-	-	0.51(F Violet)	-	0.51(Violet)	0.51(Violet)
0.55(L Green)	-	0.55(F Violet)	-	-	-
-	0.58 (L Green)	-	-	-	-
-	-	-	-	0.60(L Violet)	0.60(L Violet)
0.63(L Green)	-	-	-	-	-
-	-	0.66(F Greenish Blue)	0.66(F D Greenish Blue)	0.66(L Violet)	-
0.72(L Green)	0.72(L Green)	-	-	0.72(Violet)	-
-	-	-	-	0.73(Violet)	-
-	-	0.75(L Violet)	0.75(F L Red)	-	-
-	-	-	-	0.80(L Violet)	-
-	-	0.83(F L Violet)	0.83(F L Red)	0.83(Violet)	-
-	-	-	-	0.87(L Violet)	-
-	-	0.92(F L Violet)	0.92(F L Violet)	0.92(L Violet)	-
-	-	-	-	0.94(L Violet)	-

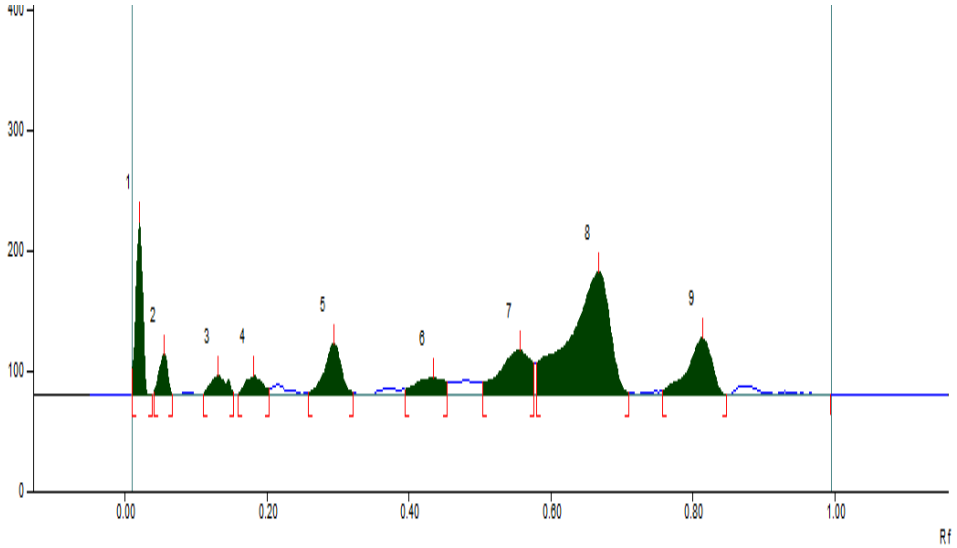
***L-Light, D-Dark, F-Fluorescence**

Fig 4. Densitometric scan At 254nm



Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	32.2 AU	0.02 Rf	66.9 AU	5.46 %	0.04 Rf	2.0 AU	610.5 AU	2.31 %
2	0.05 Rf	1.6 AU	0.07 Rf	48.1 AU	3.92 %	0.09 Rf	0.4 AU	588.7 AU	2.23 %
3	0.13 Rf	2.3 AU	0.14 Rf	29.8 AU	2.43 %	0.16 Rf	0.1 AU	370.8 AU	1.40 %
4	0.17 Rf	0.3 AU	0.20 Rf	22.8 AU	1.86 %	0.20 Rf	18.1 AU	288.4 AU	1.09 %
5	0.21 Rf	18.2 AU	0.23 Rf	38.4 AU	3.14 %	0.25 Rf	0.6 AU	764.3 AU	2.89 %
6	0.26 Rf	0.8 AU	0.30 Rf	97.4 AU	7.95 %	0.31 Rf	60.0 AU	2038.2 AU	7.71 %
7	0.32 Rf	60.6 AU	0.36 Rf	229.8 AU	18.75 %	0.40 Rf	41.1 AU	6937.3 AU	26.24 %
8	0.40 Rf	41.4 AU	0.43 Rf	116.6 AU	9.51 %	0.45 Rf	95.7 AU	2571.4 AU	9.73 %
9	0.45 Rf	96.7 AU	0.46 Rf	121.7 AU	9.93 %	0.49 Rf	24.4 AU	2374.8 AU	8.98 %
10	0.49 Rf	25.2 AU	0.53 Rf	187.4 AU	15.30 %	0.57 Rf	1.7 AU	4345.7 AU	16.44 %
11	0.61 Rf	3.2 AU	0.64 Rf	26.5 AU	2.16 %	0.66 Rf	0.4 AU	435.7 AU	1.65 %
12	0.69 Rf	21.0 AU	0.72 Rf	60.1 AU	4.90 %	0.75 Rf	18.5 AU	1391.6 AU	5.26 %
13	0.78 Rf	15.8 AU	0.81 Rf	62.7 AU	5.12 %	0.84 Rf	10.5 AU	1350.6 AU	5.11 %
14	0.84 Rf	10.6 AU	0.88 Rf	67.3 AU	5.49 %	0.91 Rf	0.4 AU	1493.7 AU	5.65 %
15	0.92 Rf	0.1 AU	0.94 Rf	50.0 AU	4.08 %	0.97 Rf	0.1 AU	879.0 AU	3.32 %

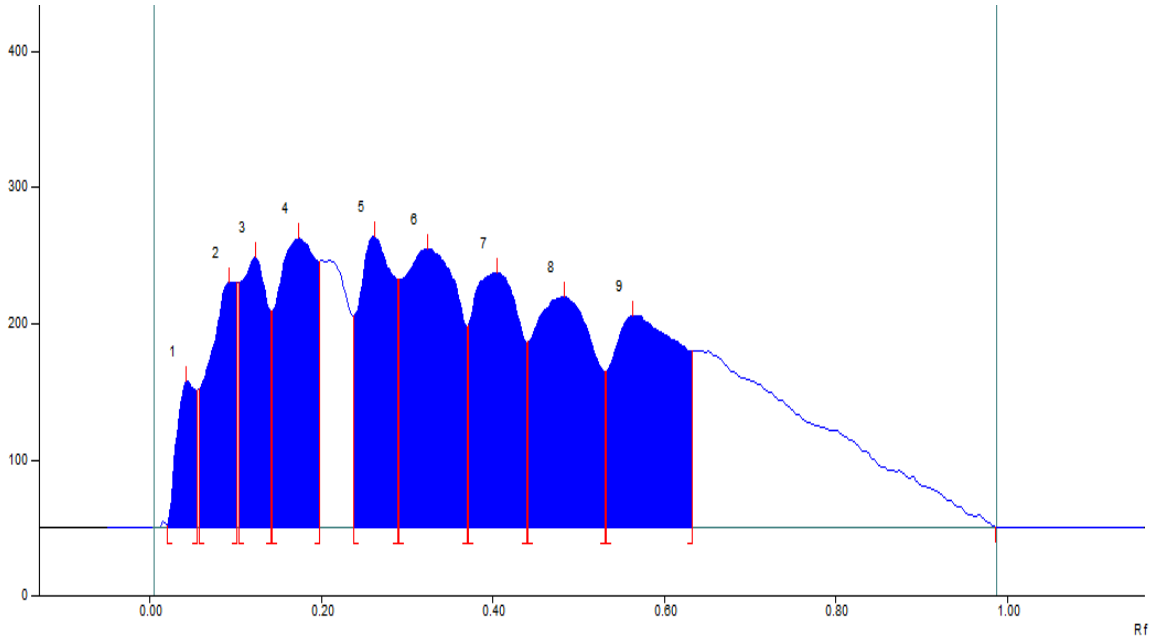
Fig 4.a *Yashtimadhu churna* (8 µl)



Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	21.1 AU	0.02 Rf	143.9 AU	31.94 %	0.04 Rf	0.0 AU	947.8 AU	10.32 %
2	0.04 Rf	1.4 AU	0.06 Rf	33.6 AU	7.45 %	0.07 Rf	0.2 AU	289.3 AU	3.15 %
3	0.11 Rf	0.3 AU	0.13 Rf	15.6 AU	3.45 %	0.15 Rf	1.1 AU	254.5 AU	2.77 %
4	0.16 Rf	0.3 AU	0.18 Rf	14.9 AU	3.32 %	0.20 Rf	4.8 AU	252.1 AU	2.74 %
5	0.26 Rf	1.9 AU	0.30 Rf	42.5 AU	9.43 %	0.32 Rf	1.9 AU	696.1 AU	7.58 %
6	0.40 Rf	4.4 AU	0.44 Rf	14.6 AU	3.24 %	0.45 Rf	10.4 AU	404.3 AU	4.40 %
7	0.50 Rf	10.3 AU	0.56 Rf	36.6 AU	8.12 %	0.58 Rf	27.1 AU	1128.4 AU	12.28 %
8	0.58 Rf	26.6 AU	0.67 Rf	101.9 AU	22.62 %	0.71 Rf	1.9 AU	4078.6 AU	44.40 %
9	0.76 Rf	2.7 AU	0.81 Rf	46.9 AU	10.42 %	0.85 Rf	0.7 AU	1134.5 AU	12.35 %

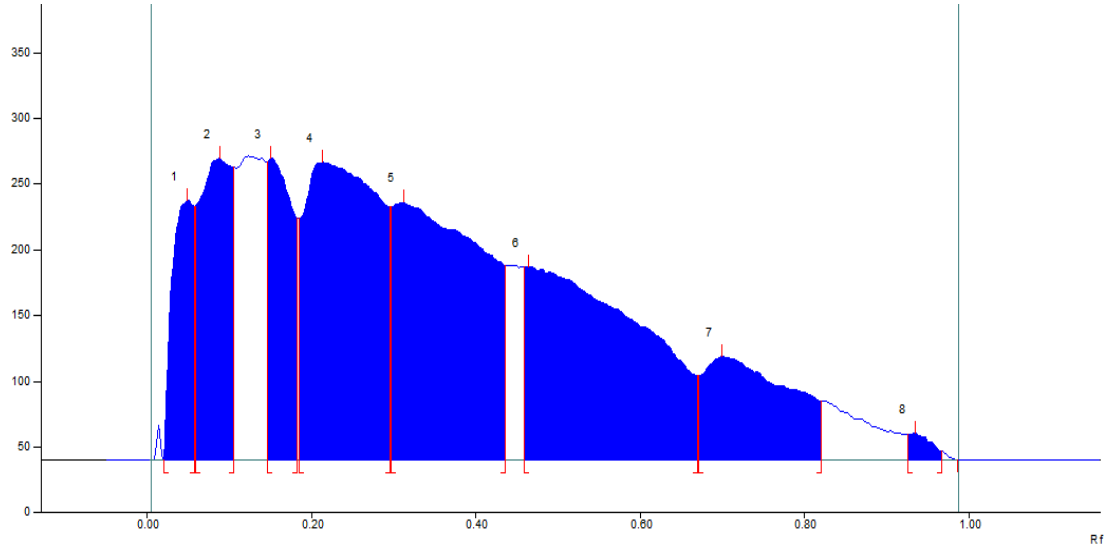
Fig 4.b *Manjishta Churna* (8 µl)

Fig 5. Densitometric scan At 254nm



Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.02 Rf	1.3 AU	0.04 Rf	107.4 AU	6.59 %	0.06 Rf	00.8 AU	1736.2 AU	3.01 %
2	0.06 Rf	101.1 AU	0.09 Rf	180.3 AU	11.06 %	0.10 Rf	80.1 AU	4106.6 AU	7.12 %
3	0.10 Rf	179.5 AU	0.12 Rf	198.9 AU	12.21 %	0.14 Rf	58.4 AU	4395.8 AU	7.62 %
4	0.14 Rf	158.7 AU	0.17 Rf	212.5 AU	13.04 %	0.20 Rf	95.3 AU	6873.8 AU	11.91 %
5	0.24 Rf	155.4 AU	0.26 Rf	213.3 AU	13.09 %	0.29 Rf	81.6 AU	6306.2 AU	10.93 %
6	0.29 Rf	181.8 AU	0.32 Rf	204.6 AU	12.55 %	0.37 Rf	47.2 AU	9411.9 AU	16.31 %
7	0.37 Rf	148.4 AU	0.41 Rf	187.2 AU	11.49 %	0.44 Rf	35.9 AU	7361.6 AU	12.76 %
8	0.44 Rf	136.1 AU	0.48 Rf	169.6 AU	10.41 %	0.53 Rf	14.5 AU	8609.6 AU	14.92 %
9	0.53 Rf	114.9 AU	0.56 Rf	155.7 AU	9.55 %	0.63 Rf	29.4 AU	8900.7 AU	15.43 %

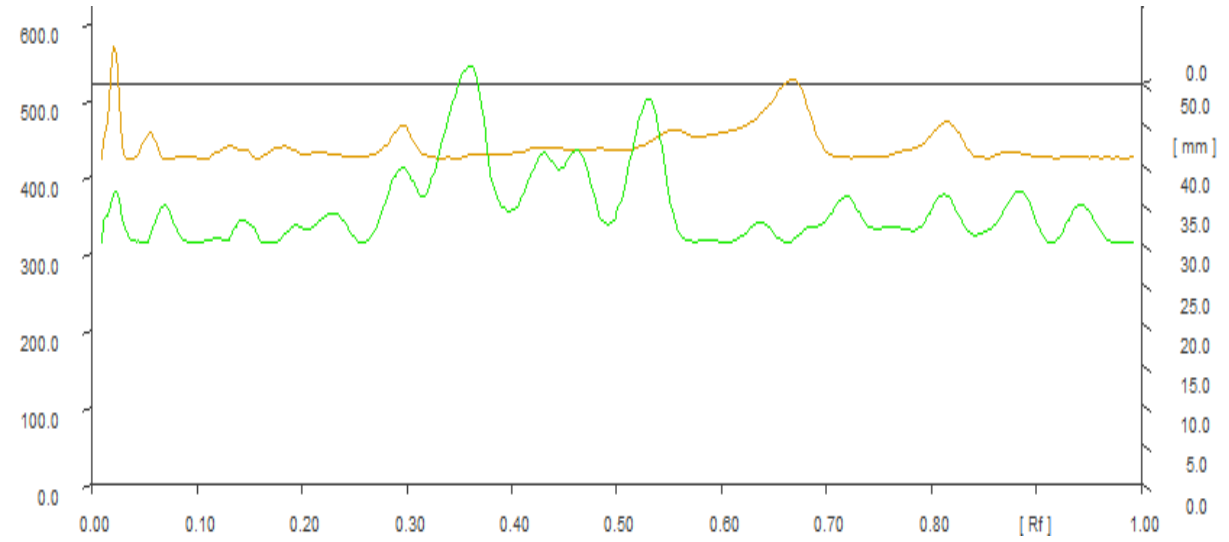
Fig 5a.*Yashtimadhu churna* (8 µl)



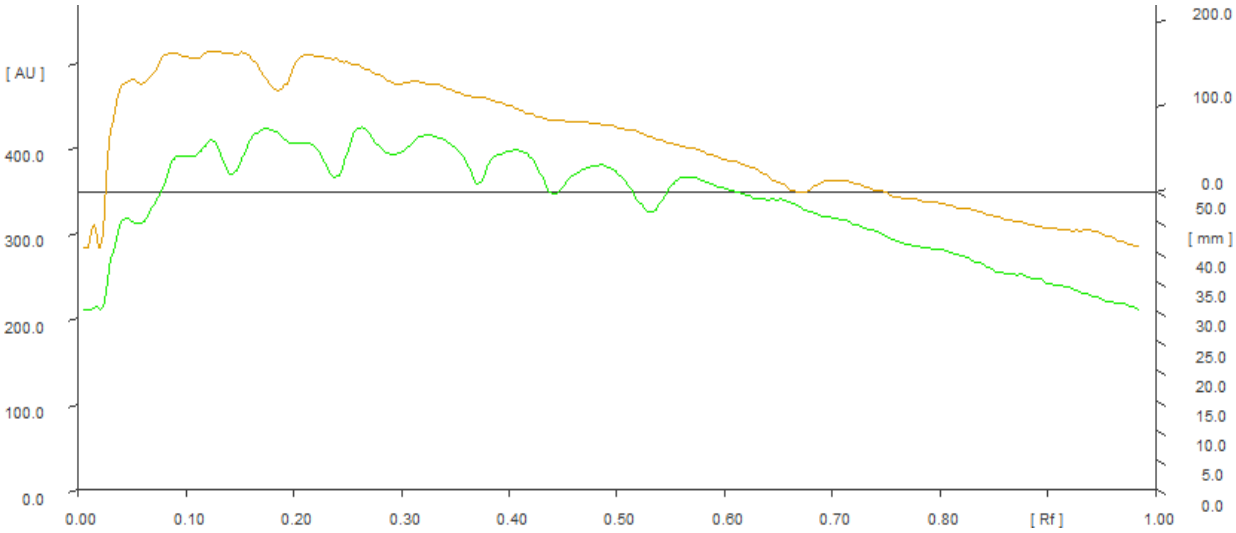
Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.02 Rf	0.3 AU	0.05 Rf	197.2 AU	14.89 %	0.06 Rf	92.4 AU	3662.9 AU	5.50 %
2	0.06 Rf	192.8 AU	0.09 Rf	229.2 AU	17.31 %	0.11 Rf	22.3 AU	6521.0 AU	9.79 %
3	0.15 Rf	226.6 AU	0.15 Rf	229.6 AU	17.34 %	0.18 Rf	83.5 AU	5074.0 AU	7.62 %
4	0.19 Rf	183.5 AU	0.21 Rf	226.3 AU	17.09 %	0.30 Rf	92.4 AU	14782.1 AU	22.20 %
5	0.30 Rf	192.8 AU	0.31 Rf	196.1 AU	14.81 %	0.44 Rf	47.7 AU	15246.2 AU	22.90 %
6	0.46 Rf	146.9 AU	0.46 Rf	147.0 AU	11.10 %	0.67 Rf	63.9 AU	15008.3 AU	22.54 %
7	0.67 Rf	64.1 AU	0.70 Rf	78.5 AU	5.92 %	0.82 Rf	44.7 AU	5882.7 AU	8.84 %
8	0.93 Rf	19.8 AU	0.94 Rf	20.3 AU	1.53 %	0.97 Rf	6.7 AU	398.7 AU	0.60 %

Fig 5.b ManjishtaChurna (8 µl)

Fig 6.3-D Display of All the samples



At 254 nm



At 366 nm

DISCUSSION

Skin lightening is not only a psychological and social issue, but also related to general health issue that needs to be addressed with some interventions. As tyrosinase inhibition is still the most sought after mechanism of skin lightening, herbs having such property will show promise as depigmenting agents.[1] There are two main types of melanin that determine skin tone viz. Eumelanin and Phaeomelanin. Individuals with darker skin tones have mostly eumelanin as compared to phaeomelanin and vice-versa.[12]

It is common to have many plant ingredients in a single herbal formulation. Due to the complex nature and variability of the constituents, herbal preparations are likely to have variations right from the stage of collection of raw materials. In the past, due to the absence of a standard reference for identification, it was difficult to establish the quality control measures for polyherbal formulations. However, nowadays, efforts have been made so that herbal preparations comply with the consistent standards through modern analytical techniques. [13]

The powder microscopy and phytochemical tests carried out in the present study serve as the preliminary tests for the standardization of the formulation. Tests such as tests for alkaloids, steroids, saponins, tannins, flavonoids, phenol, coumarins, triterpenoids, carboxylic acid, resin, quinone, HPTLC, results of HPTLC photo documentation, the unique Rf values, densitometric scan and densitogram obtained at different wavelengths can be used as fingerprint to identify both the herbal drugs, *Yashtimadhu Churna* (*Glycirrhizia glabra*) powder and *Manjishtha churna* (*Rubia Cordifolia*) powder and also to be used as complexion promoting drugs.

Studies on *Yashtimadhu churna* have also shown that the role of *G. glabra* on skin is mainly attributed to its antioxidant activity of phytochemicals namely triterpene, saponins (Glycyrrhizin-salts of glycyrrhizic acid) and flavonoids. Glycyrrhizetic acid controls the secretion of melanin in skin and it has the effect of reducing dark pigmentation and making the complexion fairer.[1]

Manjisthachurna holds the reputation of a very good skin care herb as is used to make the complexion even and lighten dark spots.[14] Ayurvedic texts enumerate its qualities to be: *Varṇya*, *Raktaprasādaka*, *Raktashodhaka* (blood purifier). Chemically, it contains glucosides known as Manjisthin and Purpurine, along with resins, lime salts and colouring agents.1Methanolic extract of this herb has been reported to show 14.80% mean inhibition of tyrosinase activity thereby acting as skin whitening agent.[15]

In the present study, both the *Yashtimadhu Churna* (*Glycirrhizia glabra*) or the powder and *Manjishtha churna* (*Rubia Cordifolia*) or the powder shows that they are endowed with various biological properties and efforts have been made here to provide scientific data on the same and Hence these drugs can be used as the standard *Varnya dravya* or the complexion promoting drugs.

CONCLUSION

Accurate Ayurvedic drug standardization is a big challenge as it requires rational approach and in this regard, fundamental aspects of Ayurvedic drug should be preserved. Main drawback in Ayurvedic drug standardization is the identification of biological source of the drug. The active constituent may vary according to geographical source of the drug and it may not be easy to standardize drug chemically. The results obtained through this study were quick, reproducible and could be used for routine monitoring of raw material. The parameters used in this work ensure the quality control of raw material, processed powder. Both *Yashtimadhu churna* (powder) and *Manjishtha churna* (powder) were standardized as per standard testing protocol through powder microscopy, preliminary phytochemical analysis and HPTLC.HPTLC photo documentation, R_f values and densitometric scan of the given samples were also recorded.

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