



## Research Article

## PHYSICO CHEMICAL ANALYSIS OF VRANASHODHANAHARA TAIL AND DOORVADI TAIL- A COMPARATIVE STUDY

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### ABSTRACT

*Tail Kalpana* (Medicated Oil) is a pharmaceutical process. It comes under the *Sneha Kalpana*. It may be defined as a process where the various things like decoction, paste, milk and perfuming - substances are employed for the preparation of oleaginous medicaments (Oil and Ghee). This process ensures absorption of the active therapeutic properties of the ingredients used. The experimental test was done in laboratory, Department of Rasa Shastra & B.K., N.I.A., Jaipur and Oasis Lab., Jaipur by adopting S.O.P. and S.M.P. as laid down in the respective pharmacopeias. Physico - chemical properties of both the medicated oils and untreated oil were determined. The analysis includes as following- Specific gravity, Refractive Index, Loss on drying, Ash value, Acid Value, Saponification value, Ester value, Iodine Value, Unsaponifiable matter, thin layer chromatography and Spectrophotometry. The T.L.C. pattern of Unsaponifiable matters of medicated oils and base oil indicates the presence of active constituents. The absorption characteristic of the medicated oils and base oil were Studied as 0.08% v/v solutions of the oils in Cyclohexane were merely transparent in the visible region of the spectrum. The Spectra resembles those of Sesamin and Sesamol, the lignan Substances of Sesame oil. So it's concluded that medicated oils are better than base oil for their medicinal purpose.

### INTRODUCTION

The uses of medicated oils are as old as *Ayurvedic* system of medicine. The oils are generally prepared with *Tila tail* as a base<sup>[1]</sup>. The rationality behind taking oil as a base is presumably to extract lipid soluble active fractions from the ingredients into the oil. By keeping this in view the *Vranashodhanahara Tail* & *Doorvadi Tail* were prepared with an objective to find out the difference between these two medicated oils. Physical - chemical properties of both the medicated oils and untreated oil were determined.

### AIM AND OBJECTIVES

- ❖ Experimental study of *Vranashodhanahara tail* and *Doorvadi tail* for their physical and chemical analysis.
- ❖ Elaborate the methods of physical and chemical analysis of medicated oil.

### MATERIAL AND METHODS

The trial drugs *Vranashodhanahara tail* contains *Tila Tail*, *Tila Beej* (*Seasamum indicum*), *Nimba*

*Patra Swarasa* (*Azadirachta indica*) *Haridra* (*Curcuma longa*) and *Trivrit* (*Operculina terpathum*) where as the *Doorvadi tail* consists of *Tila Tail*, *Kampillaka* (*Mallotus philippinensis*), *Daruharidra* (*Berberis aristata*) and *Doorva Swarasa* (*Cynodon dactylon*). The formulations *Vranashodhanahara Tail* (Pharmacopeia Govt. of A.P.) and *Doorvadi Tail* (B.R. 47/79-80) were prepared and test was done in laboratory, Department of Rasa Shastra & B.K., N.I.A., Jaipur, by adopting S.O.P. and S.M.P. as laid down in the respective pharmacopeias.

**Experimental study:** The analysis includes determination of the following test<sup>[2]</sup>.

1. Specific gravity
2. Refractive Index
3. Loss on drying
4. Ash value
5. Acid Value
6. Saponification value
7. Ester value
8. Iodine Value

9. Unsaponifiable matter
10. Thin layer chromatography
11. Spectrophotometry

**Specific Gravity:** The specific gravity of a liquid is weight of a given volume of a liquid at a specific temperature compared with the weight of an equal volume of water at the same temperature. The presence of dissolved substances in oil is expected to change its specific gravity, therefore it is considered to be an important parameter for analyzing medicated oils.

**Method:** A cleaned 25ml capacity picnometer filled with water up to the mark at ambient temperature and its weight was taken. The picnometer was next filled with the sample upto the mark at the same temperature and again the weight was noted. The specific gravity is determined by dividing the weight of the sample with the weight the water expressed in grams. All the weights were taken in air. (Table no. 1).

**Refractive index:** The refractive index of a substance is the ratio of the sine of the velocity of the light in vacuum to its velocity in the substance. It may also be defined as the ratio of the angle of incidence to the sine of the angle of refraction.

**Method:** Refractive index was measured in Abbes' refractometer. The temperature of refractometer was maintained at 40°C by circulating water. The readings were taken after half a minute of pouring a drop of oil sample between the prisms. (Table no. 1).

**Loss on drying:** Loss on drying is the loss in weight percent w/w the amount of volatile matter of any kind that can be driven off under the condition specified. The test was carried out on 1.0 g of the sample, previously mixed well.

**Method:** First weighed a glass - stoppered, shallow weighing bottle that had been dried for 30 minutes. The sample taken into the bottle, covered it and accurately weighed the bottle along with the contents. The sample was distributed as evenly as practicable by gentle sidewise shaking to a depth not exceeding 10mm. The loaded bottle was placed in the drying chamber (oven) and the stopper was removed and left it in the chamber. The sample was dried for one hour at 110°C. After completion of drying, opened the chamber, closed the bottle promptly and allowed it to come to room temperature. Finally the bottle was weighed and the values were calculated. (Table no. 1).

**Ash Value:** Ashing is the process of mineralization for preconcentration of trace substances prior to chemical analysis. Ash is the name given to all non- aqueous residues that remain after sample is burned, which consists mostly of metal oxides.

**Method:** Heat a silica crucible to red hot for 30 minutes, allow it to cool in a desiccators and weighed. Accurately about 1g of the substance being examined and evenly distribute in the crucible. Dried at 100° to 105° for 1 hour and ignite to constant weight in a

muffle furnace at 600° ± 25°. Allowed the crucible to cool in desiccators was after each ignition. Care was taken the material should not catch fire at any time during the procedure. If after prolonged ignition a carbon - free ash obtained, exhaust the charred mass with hot water, collect the residue on an ash less filter paper incinerate the residue and filter paper until the ash is white or nearly so, add the filtrate, evaporate to dryness and ignite at a temperature not exceeding 450°. It was calculated the percentage of ash with reference to the air dried drug. (Table no. 1).

**Acid Value:** The acid value of oil is defined as the number of mg. of potassium hydroxide required to neutralize the free fatty acid present in 1 g. of the sample.

For estimation of the acid value about 10 g. of the sample oil was accurately weighed in a 250 ml flask and dissolved in a 50 ml mixture of equal volumes of alcohol and solvent ether and triturated with N/10 potassium hydroxide using 1 ml of phenolphthalein solution as indicator. The appearance of faintly pink color was end point. (Table no. 1).

Acid value was calculated with the following formula.

$$\text{Acid value} = \frac{N \times 5.61}{W}$$

N = the number of ml of N/10 Potassium hydroxide required.

W = the weight in grams of the substance

**Saponification value:** Saponification value is the number of milligrams of potassium hydroxide required to neutralize the free fatty acids resulting from the complete hydrolysis of 1 g of the substance. Accurately weighed 2 g. substance was taken in a 200 ml. flask refluxed with 25 ml. of 0.5 N alcoholic solution of potassium hydroxide and boiled for 30 minutes with frequently rotating the contents. The 1 ml solution of phenolphthalein was added and the excess of alkali was triturated with N/2 hydrochloric acid and the number of ml of acid (titrant) required was measured (a). (Table no. 1).

A blank test was repeated under the same condition, without the substance being tested. The volume of titrant in ml (b) was recorded. And the Saponification value was calculated from this formula.

$$\text{Saponification value} = \frac{(b-a) \times 28.05}{W}$$

W = weight in grams of the substance

**Ester Value:** The ester value is the number of mg. of potassium hydroxide required to saponify the esters (neutralizing the acids resulting from the complete hydrolysis of fat and not the free fatty acids present in it) present in 1 g. of the substance. (Table no. 1).

The values can be obtained by subtraction of acid value from Saponification value.

Ester value = Saponification value - Acid Value

**Iodine value:** Iodine value of a substance is the number of grams of iodine absorbed by 100 g. of the substance. Oils contain both saturated and unsaturated glycerides and iodine value is a measure of the degree of unsaturation of oil.

About 2 g. of oil was accurately weighed, placed in a dry iodine flask and dissolved with 20 ml carbon tetrachloride. In this solution 25 ml of pyridine bromide solution was added and allowed to stand for ten minutes in the dark place. And then 15 ml of potassium iodide (10%) was added in it. The stopper and the sides of the flask were rinsed with 100 ml of water and titrated with 0.1N sodium thiosulphate, using starch solution added towards the end of the titration, as indicator. The number of ml of the thiosulphate required was termed '(a)'. At the same time a test exactly in the same manner was carried out, but without the substance being tested, and the number of ml. of 0.1 N sodium thiosulphate required was regarded as blank titer '(b)'.

The iodine value was calculated using the formula.

$$\text{Iodine value} = \frac{(b-a) \times 0.01269 \times 100}{W}$$

W = Weight in grams of the substance.

#### UNSAPONIFIABLE MATTER

Oils are glycerides, which produce soaps and glycerol on alkali hydrolysis. In addition to glycerides, oils also contain sterols and other related fat - soluble compounds, which do not form soaps with alkali. In medicated oil is important because it is assumed that the active ingredients from the vegetable drugs pass into the oil under the condition when it is prepared.

About 2 gm of accurately weighed oil, taken in a 250 ml flask, was refluxed with 40 ml N/2 alcoholic potassium hydroxide on water bath for one hour, with frequent swirling of the contents. The contents of the flask were transferred into a separatory funnel by means of 100 ml water and the warm liquid was extracted by shaking vigorously with three successive quantities, each of 50 ml of solvent ether. The flask used for reflux was washed with the first quantity of solvent ether and all the three extracts were mixed together in a separatory funnel and washed with 20 ml of water without violent shaking. The liquids were allowed to separate and the aqueous layer was removed. The ethereal layer was

washed again by shaking vigorously with two successive quantities, each of 20 ml of water and then treated three times with 20 ml of N/2 aqueous potassium hydroxide, shaking vigorously on each occasion, each treatment being followed by washing with 20 ml of water.

Finally the ether layer was washed with successive quantities, each of 20 ml of water until the aqueous washing failed to develop pink colour with phenolphthalein solution. The ethereal extract was transferred to a weighed flask washing out the separator with solvent ether. The ether was distilled off and 3 ml of acetone was added in the flask. The solvent was completely removed from the flask. The flask was dried to constant weight at 80°C for 30 minutes. And the percentage of Unsaponifiable matter was calculated. (Table no. 1).

#### THIN LAYER CHROMATOGRAPHY<sup>[3]</sup>

The Unsaponifiable matter of plain & medicated oils were chromatographic over TLC plates to ascertain whether any plant constituent have been transferred into the medicated oils or not. The silica gel - GF - 254 plates were developed with mobile phase. Ethyl acetate: Benzene (2:8). At the end of the process the plates were visualized for spots by naked eye, exposing to Iodine vapors and under UV light (at 254 nm). (Table no. 2).

#### SPECTROPHOTOMETRY<sup>[4]</sup>

Absorption spectrometry is the measurement of the absorption of electromagnetic radiation of definite and narrow wavelength, range by molecules and atoms of a chemical substance or a sample. These techniques frequently employed in pharmaceuticals include ultra-violet, visible, infrared and atomic absorption spectroscopy. For many pharmaceutical substances, measurements can be made in the ultra violet and visible regions of the spectrum with greater accuracy and sensitivity in this research work the ultra violet spectroscopy was done. The wavelength range was available for this measurement extends from 185 nm to 380 nm.

The absorption characters of the oils in visible and U.V. region of the spectrum were studied. 0.08% v/v solutions of the oils in cyclohexane were merely transparent in the visible region of the spectrum. (Table no. 3).

**Table 1: Showing Physico-Chemical Standards of Vranashodhanahara Tail & Doorvadi Tail**

S.No.	Parameter	Sample P	Sample V	Sample D
1.	Colour	Pale Yellow	Greenish dark	Blackish Brown
2.	Smell	Pleasant	Characteristic	Characteristic
3.	Appearance	Viscous	Viscous	Viscous
4.	Touch	Oily	Oily	Oily
5.	Clarity	Clear	Not Clear	Clear
6.	Taste	Bitter	Bitter	Slight Bitter
7.	Opalescence	Transparent	Not Transparent	Not Transparent
8.	Sp. Gravity	0.9140	0.9169	0.9169

9.	Refractive Index	1.4690	1.4685	1.4710
10.	Loss on Drying	0.031%	0.176%	0.208%
11.	Ash Value	0.027% w/w	0.033% w/w	0.047% w/w
12.	Acid Value	1.255	6.69	1.443
13.	Saponification Value	191.32	182.84	187.07
14.	Ester Value	190.06	176.15	185.62
15.	Iodine Value	124.91	108.94	107.68
16.	Unsaponifiable Matter	0.65% w/w	0.729% w/w	0.85% w/w

**Table 2: Showing TLC of sample (R<sub>F</sub> Value)**

Sample	Visualization → Mobile Phase ↓	Naked Eye	Ultra Violet
			254 nm
<i>Vranashodhanahara Tail</i>	Benzene : Ethyl Ocetate (2 : 8)	----	0.97 and 0.86
<i>Tila tail</i>	Benzene : Ethyl Ocetate (2 : 8)	-----	No Spot Observed
<i>Doorvadi tail</i>	Benzene : Ethyl Ocetate (2 : 8)	-----	0.94

**Table 3: Showing absorption spectrometry**

1	<i>Vranashodhanahara Tail</i> – Light absorption is maximum at	286 nm =0.283 407 nm=0.226
2	<i>Doorvadi Tail</i> – Light absorption is maximum at	275 nm = 1.276 343 nm =0.453
3	<i>Tila Tail</i> – Light absorption is maximum at	270 nm =0.160 281 nm = 0.139

 Control-P: Tila tail, Sample- V: *Vranashodhanahara Tail*, Sample-D: *Doorvadi Tail*

## RESULTS AND DISCUSSION

The above data of specific gravity show that rise in the value in sample 2 and 3 in comparison to sample 1, may be due to presence of moisture and fat soluble active constituents incorporated through the drugs. The refractive index is highest in sample 3. The other sample was relatively very close even on heat. The values of loss in drying were differing with each other. The higher value observed in sample 3 might be the high value in sample 3 might be due to *Kalka dravya*. The ash values are very close in sample 1 & 2. The sample 3 was higher than other two. The acid value of base oil (*Tila tail*) is within the worth of official standard, whereas the value was found high in *Vranashodhanahara Tail*, may be due to presence of acidic constituents from *Tila*, *Haridra*, *Trivrit* etc. The value was found lower in the sample 1. In saponification Value, the data showed that the base oil was found higher value. The value decreased in finished products may be due to removal of moisture and other non fatty volatile compounds. The ester value of base oil was found high. The value decreased in finished products may be due to removal of moisture and other non fatty volatile compounds. The iodine value of base oil was found high. The value was decreased due to heating process obviously breakdown or oxidation across the double bond of unsaturated fat. Unsaponifiable matter is the non fatty matter, which are compounds other than glycerides and fatty acids that remain soluble in fats. On hydrolysis of fats and oils, the glycerides and free fatty acids are converted into soap and glycerin and Unsaponifiable matter remain suspended in soap solution, which can be extracted with organic solvent like ether. In order to ascertain whether any plant constituent other than fats

has been transferred to the medicated oil from the drug during the processing. The Unsaponifiable matters of *Tila tail* as well as of medicated oils were chromatographic over TLC plates. The plates were developed with Benzene and ethyl acetate (2:8) and the chromatograms were sprayed with L.B. reagent for visualization of spots. The spot of the medicated oil were found to be identical with those of sesame oil. It was thus not possible to detect the presence of any additional constituent in the Unsaponifiable matter. It was inferred that active constituents of any transferred to the medicated oil is present only in traces and not detectable by TLC analysis without prior fractionation with large quantity of materials<sup>[5]</sup>. The test of absorptive spectrometry, in the UV region, all the samples showed two different absorption maxima at 270 nm and 407 nm. This spectrum resembles that of sesamin and sesamol, the lignans known to be present in sesame oil. (Table no. 1 to 3)

## CONCLUSION

*Vranashodhanahara tail* and *Doorvadi tail* are popular oil preparations for *Varnashodhana* and *Ropana* in Ayurveda. The oil preparations include both fixed and essential oils. It was observed that the degree of heat and constituent of the drugs affects the physical and chemical constants such as Specific gravity, Ash value etc. in individual cases<sup>[6,7]</sup>.

The T.L.C. pattern of unsaponifiable matters of medicated oils and base oil which indicates the presence of active constituents, if any transferred from the drugs to the oil will be in traces not detectable by T.L.C. analysis without prior fractionation involving large quantity of material.

The absorption characteristics of the medicated oils and base oil were studied 0.08% v/v solutions of the oils in Cyclohexane were merely transparent in the visible region of the spectrum. The spectrum resembles those of Sesamin and Sesamolin, the lignan Substances of Sesame oil. So it is concluded that medicated oils are better than base oil for their medicinal purpose.

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SHOWING THE PREPARATION OF DOORVADI TAIL



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