Standardisation of Talisadi Taila- A Noble medicine having wound healing property

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Abstract

Taila Kalpanas (Medicated Oils) are the integral part of the Ayurvedic treatment. Tailas are used in the treatment of diseases explained in Astangas of Ayurveda for both bahya and abhyantara chikitsa (Paana, Nasya, Basti, Abhyanga). Talisadi Taila which is mentioned in Ashtanga Hridaya for its best Vrana-ropaka (Wound Healing effect). Such marvelous preparations remained untouch or not introduced into the Vaidya community till date. Talisadi taila has been prepared as per the Standard Operating Procedures (SOP) which have been mentioned in Sharangadhara Samhita for the preparation of aushadhisiddha tailas,. The studies on the identity, purity and quality of guanine taila will enhance information in checking the adultration.. Efforts have been made to unmask such safe potent medicated oil for its wound healing and establishing the standard parameters for its identity, purity is concerned.

Key Words: Talisadi Taila, Wound, Adultration, SOP, Abhyanga.

Introduction:

Sneha Kalpana (Medicated oil preparations), one of the important secondary dosage form in Ayurvedic pharmaceutics have a broad spectrum use in different medical conditions. Various crude oils are mostly associated with rancidity factors(amadosha), those are effectively removed and simultaneously therapeutic quality is enhanced by the ancient ayurvedic pharmaceutical techniques called taila Taila Murchhana. Such murchhita tila taila was used for the preparation of Talisadi Taila. Talisadi Taila which is mentioned in Ashtanga Hridaya for its best Vrana-ropaka (Wound Healing effect). Such marvelous preparation remained untouch or limited use or not introduced into the Vaidya community till date. The ingredients which are used in the preparation of Talisadi taila possess therapeutically proven wound healing
properties like, Agaru, Chandana, Madhuka, Haridra, Daruharidra, Padmabeeja etc.

Aims and objectives:
1) To prepare Talisadi Taila as per SOPs laid down in the classics.
2) To conduct Physico-chemical analysis of Talisadi Taila.
3) To establish standard parameters for Talisadi Taila.

Materials and Methods:
Murchita Tila Taila & ingredients of Talisadi taila were used for the preparation of Talisadi taila. And the sample was subjected for physico-chemical analysis like refractive index, specific gravity, Saponification value, acid value and Iodine value etc.

Materials:
The following materials were used for the preparation of Talisadi Taila.

METHOD OF PREPARATION OF TALISADI TAILA –

Apparatus Required:
Weighing machine, wide mouthed vessel, Khalwayantra, Darvi, clean cloth, Chullika, Kalkanishpeedana yantra

Ingredients & their Quantities:

<table>
<thead>
<tr>
<th>S. NO.</th>
<th>INGREDIENTS</th>
<th>QUANTITY</th>
</tr>
</thead>
<tbody>
<tr>
<td>01.</td>
<td>Talisa Patra</td>
<td>180 gm.</td>
</tr>
<tr>
<td>02.</td>
<td>Padmaka</td>
<td>180 gm.</td>
</tr>
<tr>
<td>03.</td>
<td>Jatamansi</td>
<td>180 gm.</td>
</tr>
<tr>
<td>04.</td>
<td>Harenuka</td>
<td>180 gm.</td>
</tr>
<tr>
<td>05.</td>
<td>Aguru</td>
<td>180 gm.</td>
</tr>
<tr>
<td>06.</td>
<td>Chandana</td>
<td>180 gm.</td>
</tr>
<tr>
<td>07.</td>
<td>Haridra</td>
<td>180 gm.</td>
</tr>
<tr>
<td>08.</td>
<td>Daruharidra</td>
<td>180 gm.</td>
</tr>
<tr>
<td>09.</td>
<td>Kamalabeej</td>
<td>180 gm.</td>
</tr>
<tr>
<td>10.</td>
<td>Ushira</td>
<td>180 gm.</td>
</tr>
<tr>
<td>11.</td>
<td>Yashtimadhu</td>
<td>180 gm.</td>
</tr>
<tr>
<td>12.</td>
<td>Murchita Tila Taila</td>
<td>1.8 liters</td>
</tr>
<tr>
<td>13.</td>
<td>Jala</td>
<td>7.2 liters</td>
</tr>
</tbody>
</table>

Method of preparation:
Oil was taken in a wide mouth iron vessel and kept on mandagni till getting nisphena then it was removed from agni and allowed to cool. Mentioned proportion of ingredients was added and water was also added to it. The whole mixture was subjected to mandagni till taila siddhi lakshanas were appeared. After observing the taila siddhi lakshanas, removed it from agni, allowed to cool, filtered, and collected in a clear container.
Varna - Yellowish Brown Quantity of taila taken – 1.8 liters
Gandha-Pleasant Quantity of preparation – 1.6 liters
Precautions Taken:

- Mandagni was maintained throughout the procedure.
- Kalka was added only after heating the oil, and it was added little by little to avoid the spillage of the oil.
- The chronology of addition of Kalka Dravyas was maintained as mentioned in the procedure.
- Continuous stirring was carried to avoid sticking of Kalka to bottom of pan and carbonization.
- Sneha Siddhi Lakshanas were observed repeatedly and it was confirmed by testing the varti made out of Kalka.
- Care was taken to filter the kalka in warm state itself in order to reduce the loss.

Observations:

- Colour of Taila was changed into Yellowish brown.

The following materials are taken for the analytical study.

1. Talisadi taila was prepared with Murchita Tila Taila.
2. Electric hot plate with magnetic stirrer.
3. Heating Mantle.
4. Abbe’s Refractometer
5. Dropper
6. Specific Gravity(Pycnometer, 25ml capacity)
7. Weighing balance
8. 0.5N Alcoholic KOH solution and 0.5N HCL solution
9. Reflux condenser
10. Water Bath
11. Titration indicator (phenolphthalein)
12. 0.1N NaOH solution
13. Solvent Ether.
14. Sodium Hydroxide
15. Measuring cylinders
16. Pippete
17. Burrate
18. Beaker
19. Round bottom flask
20. Funnel

Methods:

Determination of Refractive Index:

The refractive index of a substance is the ratio of the velocity of light in vacuum to the velocity of light in the substance.

1. At first the mirror of the Abbe’s Refractometer was adjusted to 45 c. Then while sample was inserted in the prism box by using a thin dropper.
2. Various color bands were observed in the right eye piece. Color bands were removed with the help of compensator knob in such a way that only the black and white portion should be seen in the black and white portion should be seen in the right eye piece.
3. The black and white portion are adjusted to the cross wire with the help of lever. Finally the reading was
noted on the scale through left eye piece.

**Determination of Specific Gravity:**

Specific gravity of a substance is the weight of the substance in grams at a specific temperature compared with the weight of the same volume of water in grams at a same temperature.

1. A clean and dried 25ml capacity of specific gravity bottle (picnometer) was weighed empty. Then it was filled with water and weighed at room temperature.

2. Again the bottle was cleaned and dried then filled the oil sample up to the mark and weighed at the same temperature.

3. The specific gravity was determined by dividing the weight of the sample in grams by the weight of the water in grams.

Specific gravity of the sample = Weight of (Oil) sample in grams/weight of same volume of water at same temperature in grams.

**Determination of Saponification Value:**

Saponification value of an oil or fat is defined as the number of milligrams of KOH required to neutralize the fatty acids resulting from the complete hydrolysis of 1 gram of sample.

1. At first 250 ml capacity of round bottom flask is filled with a reflux condenser. Then 2 gram of oil sample with 25 ml of 0.5N KOH solution was taken into the round bottom flask.

2. Then 2-3 pieces of pumice stones were put in to the same flask and the mixute was boiled on water bath at 40°C for 30 min.

3. Afterword it was taken out from water bath and 1 ml of phenolphthalein indicator was added to it. Titration was done immediately with 0.5N HCl.

4. The burrate reading was noted (a).

5. The same procedure was carried out without taking the oil sample, i.e. a blank test under same conditions and burrate reading was noted (b).

Saponification value was determined as per following formula.

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

**Determination of Iodine Value:**

The Iodine value of oil is the weight of iodine absorbed by 100 parts by weight of the same, when determined by one of the following methods.

**Method-I (Iodine Monochloride method) (Wij’s method)**

Place the sample, accurately weighted, in a dry iodine flask of 250 ml capacity, add 10 ml of carbon tetrachloride, and dissolve. (The approximate weight in gms. of the
sample to be taken may be calculated by dividing 20 by the highest expected iodine value). Add 10 ml of chloroform and 20 ml of Iodine monochloride solution insert the stopper, previously moistened with potassium iodine solution and allow to stand in a dark place at a temperature of about 17°C for thirty minutes. Add 15 ml of potassium iodine solution and titrate with N/10 sodium thiosulphate using starch mucilage as indicator. Note the number of ml required (a). At the same time carry out the operation in exactly the same manner, but without the sample being tested and note the number of ml N/10 sodium thiosulphate required (b). Calculate the iodine value from the formula.

\[
\text{Iodine value} = \frac{(b - a) \times 0.01269 \times 100}{\text{Weight of Sample in Grams}}
\]

If (b-a) is greater than b/2, the test must be repeated using a smaller quantity of the samples.

**Method-II Iodine Monobromide Method - Hanus Method.**

Iodo bromide solutions – Dissolve 13.2 grams of iodine in 1000 ml. of glacial acetic acid with the aid of gentle heat, if necessary cool the solution to 75°C and determine the iodine content in 20 ml by titration with N/10 sodium thiosulphate. Add to the remainder of the solution a quantity of bromine equivalent to that of the iodine present. Store in glass containers, protected from light.

Place the sample, accurately weighed in a 250 ml. iodine flask, add 10 ml of chloroform and dissolve (the approximate weight in gms of the sample to be taken may be calculated by dividing 20 by the highest expected iodine number) add 25 ml of Iodobromide solution, stopper the flask, and allow it to stand for 30 minutes, protected from light. Then add in the order named 1330 ml of potassium iodine solution, (16.5 percent w/v) and 100 ml of water, and titrate the liberated iodine with N/10 sodium thiosulphate using starch mucilage as indicator. Note the number of ml required (a). At the same time, carry out the operation in exactly the same manner, but without the sample being tested and note the No. of ml of N/10 sodium thiosulphate required (b).

Calculate the iodine value from the formula.

\[
\text{Iodine value} = \frac{(b - a) \times 0.01269 \times 100}{\text{Weight of Sample in Grams}}
\]

**1. Acid Value:**

The determination of Acid value is carried out on the oil extracted from
the sample by continuous extraction with Ether.

The Acid value of oil is defined as the number of milligrams of Potassium Hydroxide required to neutralize the free acid in 1 gram of sample.

**Method:**

Mix 25ml ether with 25ml Alcohol (95%) and 1ml. of 1% phenolphthalein solution and neutralize with N/10 alkali (few drops). Dissolve about 5gm. of the oil, accurately weighed in the mixed neutral solvent, and titrate with N/10 potassium (or sodium) hydroxide, shaking constantly until a pink colour which persists for 15 seconds is obtained.

The titration should preferably not exceed about 10 ml.

\[
\text{Acid value} = \frac{N \times \text{ml. of } \text{N/10 alkali used}}{\text{Weight of Sample in Grams}} \times 561
\]

The free fatty acid content is also expressed as F.F.A. calculated as oleic acid % (1ml.N/10 alkali =0.028gm. Oleic acid).

**4. Loss on Drying at 110°C:**

Loss on drying signifies the amount of residual water in the finished product. Ideally, it should be nil in case of Ghritas and Tailas.

**Procedure:** About 5 gm. of sneha is taken in a crucible, heated to liquid consistency and weighed accurately. Then it is put in furnace, heated upto 105°C for half an hour. Then it is taken out and weighed again. The percentage of difference before and after subjecting the sample to heat is considered as loss on drying at that particular temperature.

**5. Ester value:**

It is defined as number of milligrams of Potassium hydroxide required to combine with fatty acids which are present in glycerides found in 1gm sample of oil or fat. Difference between Saponification value and acid value is ester value.

\[
\text{Ester value} = \text{Saponification value} - \text{Acid value}
\]

**7. Chromatographic Techniques:**

Chromatography represents a group of methods for separating molecular mixtures that depend on the differential affinities of the solute between two immiscible phases.

**Thin Layer Chromatography (TLC)**

- Thin layer chromatography is particularly valuable for the Qualitative
Determination of small amount of impurities.

- It is a technique used to detect, separate and isolate the different chemical constituents (Active Principles) present in a sample.
- It has an adsorbent coated on a glass plate, which is the stationary phase and the solvent system used in the mobile phase. Percolation of mobile phase through the adsorbent develops the Chromatogram.

**Materials:** Pre-coated TLC plates (Silica) of thickness 0.20 mm, 20X20 cm, applicator, glass chamber, oven, solvents, spray reagents.

**Method:**
- The TLC chamber is to be perfectly cleaned and dried before use.
- The solvent is poured into the TLC chamber and the glass lid is closed, vacuum grease is smeared on the lid so that chamber becomes air tight.
- A piece of tissue paper is kept immersed in the chamber for perfect saturation of solvent system.
- The chamber was kept undisturbed for an hour to saturate it.
- Later, the pre-coated TLC plates are taken and spotted with the help of applicator, 1 cm away from sides, and 2 cm away from the base. Space of 2 cm is maintained between each spots.
- The spotted plate is then gently immersed in TLC chamber, concentrated with the solvent in such a way that the solvent had uniform Linear contact with the plate.

**Resolution Factor (Rf value) = Distance travelled by the spot \( \frac{\text{Solvent front}}{\text{Homogeneity}} \)**

**Observation & Results:**
The characteristic Physical constants like Refractive Index and Specific Gravity are useful in the determination of purity of oils.

Saponification value indicates braking down of oil into glycerol and free fatty acids by treatment with alkali. The higher Saponification value indicates the content of low molecular weight fatty acids.

Acid value normally reflects the amount of acidity which is due to free fatty acids, acid phosphates and amino acids. This acidity is neutralized by treating with alkali. This process what is known as refining of oils.

**I. Organoleptic Characters:**

- **Colour:** Reddish-brown
- **Odour:** Characteristic
- **Appearance:** Viscous Oily
- **Clarity:** Clear

**II. Physico-chemical Parameters**
Loss on drying at 110°C : Nil

Saponification value : 178.66

Iodine value : 87.25

Acid value : 2.538
Peroxide value : 2.63

Ester value : 176.122

Refractive index at 30° C : 1.4619

Specific Gravity : 0.9191

Thin Layer Chromatography
Solvent System : Toluene: Ethyl Acetate: Acetic acid:: 8: 2: 1
Spraying Agent : Anisaldehyde Sulphuric Acid

Note: No Spots were seen under visible light before and after spray in this sample.

<table>
<thead>
<tr>
<th>Rf Values</th>
<th>Color Under Long UV Before Spray</th>
<th>Color Under Long UV After Spray</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.23</td>
<td>Fluorescent Pale Yellow</td>
<td>-</td>
</tr>
<tr>
<td>0.55</td>
<td>Fluorescent Pale Blue</td>
<td>-</td>
</tr>
<tr>
<td>0.63</td>
<td>-</td>
<td>Fluorescent Yellow</td>
</tr>
<tr>
<td>0.66</td>
<td>Fluorescent Blue</td>
<td>Fluorescent Yellow</td>
</tr>
<tr>
<td>0.70</td>
<td>-</td>
<td>Pale Blue</td>
</tr>
<tr>
<td>0.73</td>
<td>Bright Blue</td>
<td>-</td>
</tr>
<tr>
<td>0.86</td>
<td>Blue</td>
<td>-</td>
</tr>
</tbody>
</table>

Discussion:
The past two decades have seen a worldwide upsurge in the use of traditional medicine (TM) and Complementary and Alternative Medicine(CAM) in both developed and developing countries. The phyto-medicinal therapy is easy to procure and administer in different pharamaceutical dosage forms like Churna, Vati, Taila, Ghrita etc. The ingredients of Talisadi Taila has been proven scientifically for their antimicrobial and wound healing property. In the present work an extensive chemical and chromatographical analysis of Talisadi taila was undertaken. The main objective of the study was to establish the standard parameters for this noble medicine which remain untouched or undisclosed to vaidya community till date. The characteristic physical and chemical parameters were carried out which includes Refractive Index, Specific Gravity, Iodine Value, Saponification value, Acid value and chromatographical study. In the above analytical findings it is observed that the characteristic Physical constants like Refractive Index and Specific Gravity are useful in the determination of purity of oils. The higher Saponification value indicates...
the content of low molecular weight fatty acids. Lower Acid value normally reflects for its refining of oil. Which enhances the shelf life of the oil.

References:


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