Pharmacognostical and Pharmaceutical evaluation of Herbo mineral Formulation: Aragvadhadi taila

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ABSTRACT

Background: Aragvadhadi taila is a Sneha Kalpana, indicated in Kushtha Roga specially in Shvitra (Vitiligo) Roga. In present study, it has been used for external application in Shvitra (Vitiligo) Roga.

Objective: Present study is aimed to look out on herbal drugs used in the preparation of Aragvadhadi taila and standardization of Pharmacognostical, Physicochemical parameters and HPTLC evaluation.

Methods: Intermediate product sample (Kalka) identification and authentication was done by pharmacognostical study i.e. morphological features, organoleptic characters and powder microscopy. Physicochemical evaluation and HPTLC was carried out of final product. Results: Pharmacognostical Study of Intermediate product sample (Kalka) showed presence of Scleroids, Spoon cells, Tannin containing cells, Cluster crystal, Rosette crystal, Oil globules, Lignified fibres etc. Pharmaceutical evaluation showed results specific gravity 0.91, Refractive Index 1.4870, Acid Value 7.12, Saponification Value 316, Iodine Value 7.58. High Performance Thin Layer Chromatography at 254nm and 366 nm results in to 8.5 and 6 spots before and after spray respectively. Conclusion: Identification, Authentication of Herbal drugs used in the preparation and Intermediate product sample (Kalka) has been done. Physicochemical evaluation has been carried out of prepared drug which is further useful for standardization of Aragvadhadi taila and another researches.

Keywords: Aragvadhadi taila, Herbo mineral, Kushtha, Pharmacognosy, Shvitra, Standardization.

INTRODUCTION

Medicated oils occupy an important section of Ayurveda pharmaceutics described under heading of Sneha Kalpana. Aragvadhadi taila is one of the Herbo mineral formulation prescribed in Ayurvedic text Chakradatta in Kushtha Chikitsa [1]. This preparation contains five herbal drugs and two arsenicals that are Manahshila (Realgar), Haratala (Orpiment). Gomutra is used as Drava dravya and Sarshapa Taila as Sneha dravya. This formulation contains Gomutra so, five days are needed for proper paka of medicated oil as per classics [2]. It is specially indicated as in Shvitra (vitiligo) Roga. Aragvadhadi taila is one of the Herbo mineral medicated oil easily prepared oil which can be used externally to treat Shvitra (vitiligo) Roga. Present study is focus on first attempt to develops quality parameters of Aragvadhadi taila on the basis of pharmacognostical, physicochemical parameters and chromatographic evaluation. Hence, there is need to scientific proof for standardization of quality parameters. The pharmacognostic and physicochemical parameters can be used for checking the adulteration and purity of drug. Therefore, the present study was designed to evaluate the physicochemical, pharmacognostical parameters and develop the TLC fingerprint profiles of Aragvadhadi taila.

OBJECTIVE OF STUDY

Present study, is aimed to look out on herbal drugs used in the preparation of Aragvadhadi taila and Standardization of Pharmacognostical, Physicochemical parameters and HPTLC evaluation. The purpose of Standardization of raw drugs and final product is to ensure therapeutic efficacy. Therefore, maintaining the quality of this product is an essential factor.

MATERIALS & METHODS

Collection, identification, authentication of raw drugs

Collection of raw materials

Herbal Kalka Dravya like Kushtha, Haridra, Durshharidra were procured from the pharmacy of Gujarat Ayurveda University, Jamnagar. Aragvadhra fruit was collected from campus of I.P.G.T & R.A, Jamnagar. Dhava (Anogeissus Latifolia) Tvaka (bark) was collected from kevadiya colony (Gujarat).
The ingredients of Aragvadhadi taila and its part used are given at Table No 1. The raw drugs were identified and authenticated by Pharmacognosy Laboratory, IPGT & RA, Gujarat Ayurved University, Jamnagar. Identification was done on basis of organoleptic characters [Table No 2,3,4], morphological features and powder microscopy of Intermediate product sample (Kalka) as per API standards for authentication. Powder of Kalka (60#) and Aragvadhadi taila was stored in well filled closed glass containers away from the light. [Figure No I,2]

![Figure 1: Powder of Kalka (60#)](image1)

![Figure 2: Aragvadhadi Taila](image2)

Table 1: Formulation composition: Aragvadhadi taila

| No. | Ingredients | Latin / English name | Part used | Proportion |
|-----|-------------|----------------------|-----------|------------|
| 1.  | Aragvadha   | Cassia fistula Linn. | Fruit pulp | 1/7 part   |
| 2.  | Dhava       | Anogeissus latifolia | Bark      | 1/7 part   |
| 3.  | Kashtha     | Saururealappia C.B. Clerk | Root | 1/7 part |
| 4.  | Haridra     | Curcuma longa Linn. | Rhizome   | 1/7 part   |
| 5.  | Daruharide   | Berberis aristata D.C | Rhizome | 1/7 part   |

| Arsenicals (Kalka)  |
|---------------------|
| 6.  Shodhitha Manahshila | Processed Red arsenic sulphide | Whole | 1/7 part |
| 7.  Shodhitha Haratala  | Processed Yellow arsenic sulphide Whole | 1/7 part |

| Kwatha Dravya       |
|---------------------|
| 8.  Gomutra         | Cow’s urine | - | 16 part |
| 9.  Sneha Dravya    | Mustard oil | - | 4 part |

Preparation of Aragvadhadi taila in Bhaishajya Kalpana Laboratory of IPGT & RA.

**Preparation of Kalka**

Shudhibha Manahshila [1] and Shudhida Haratala [14] was powdered in porcelain kharala. Aragvadha fruit pulp was collect from Aragvadha fruit. Then, it was dissolved in sufficient quantity of Gomutra and it was boil for 10 minutes. Remained dravya from Kalka Dravya were taken in yavakuta form. Then, subjected into electrical mixer grinder and sufficient quantity of Gomutra was added in it and grinded till a paste (Kalka) was prepared. Properly made bolus kept in stainless steel vessel for further process.

**Preparation of Aragvadhadi taila**

Sarshapa Taila in the mentioned quantity was taken in a stainless steel vessel and heated over mild flame (80°C for 5 min) till complete evaporation of moisture and then bolus of Kalka were added in it. After mixing of Kalka, the specified quantity of Gomutra was added and the mixture was subjected to heat. Heating was continued maintaining the temperature in between 95-100°C with continuous stirring. The mixture was left undisturbed through the night and heating was given for 5 days. Contents were stirred continuously to avoid the possibility of settling down. Heating was continued on 5th day till Sneha Siddhi Lakshana were obtained. After obtaining desired Sneha Siddhi Lakshana, the vessel was taken out from heat and oil was filtered through two folded cotton cloth in its hot stage. The prepared oil was stored in a properly labelled air tight bottle after cooling.

**Pharmacognostical Study**

Herbal Drugs used in Intermediate product sample (Kalka) was identified and authenticated by pharmacognosy department, IPGT & RA, Gujarat Ayurved University, Jamnagar. The identification was carried out on the basis of organoleptic features, morphological features and powder microscopy of Intermediate product sample (Kalka) [5, 6].

**Pharmaceutical Evaluation**

**Physicochemical Parameters**

Aragvadhadi taila was analyzed by using qualitative and quantitative parameters at Pharmaceutical Laboratory, IPGT & RA, Gujarat Ayurved University, Jamnagar. The common parameters mentioned in Ayurvedic Pharmacopeia of India [7] and CCRAS guidelines [8] i.e. Refractive index [9], Specific gravity [10], Acid value [11], Iodine value [12], Saponification value [13] were taken.

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

**Sample preparation**

0.1 mL of oil was taken and 1 ML of hexane was added. The Solution was prepared used for chromatography. Thereafter pre chromatographic derivatization was done. Alcoholic KOH (base) and thereby heated for 10-15 minutes in CAMAG TLC plate heater. Sample application was done using CAMAG Linomat 5.

HPTLC of Aragvadhadi taila was carried out using the solvent system petroleum Ether: Diethyl ether: Acetic Acid (9:1:0.1v/v). HPTLC study was performed for the normal phase separation of components of product. Post chromatographic derivatization was done with vanillin sulphuric acid spray reagents [14].

**OBSERVATIONS AND RESULTS**

Organoleptic characters: Organoleptic characters like Taste, Colour, Odour, Touch and Texture were scientifically studied are as per detailed in Table 2,3,4.
Table 2: Organoleptic characters of raw herbal materials used in formulation

| Sr. No. | Ingredient             | Colour        | Taste      | Odour         | Touch     |
|---------|------------------------|---------------|------------|---------------|-----------|
| 1       | Aragvadha fruit pulp   | Dark Blackish | Sweet      | Strong aromatic | Slight rough |
| 2       | Dhava                  | Off-white     | Sweet      | Aromatic      | Rough     |
| 3       | Kushtha                | Brownish      | Pungent    | Aromatic      | Slight rough |
| 4       | Haridra                | Yellowish     | Pungent, Bitter | Characteristic | Rough     |
| 5       | Daruharidra            | Yellowish     | Astringent, Bitter | Characteristic | Rough     |

Table 3: Organoleptic characters of Intermediate product sample except arsenicals (Kalka)

| Sr no. | Various parameters | Results        |
|--------|--------------------|----------------|
| 1      | Colour             | Yellowish brown|
| 2      | Odor               | Aromatic       |
| 3      | Taste              | Katu, Tikta, Madhura |
| 4      | Touch              | Slightly rough |
| 5      | Texture            | Paste          |

Table 4: Organoleptic characters of prepared Drug (Aragvadhadi taila)

| Sr no. | Various parameters | Results        |
|--------|--------------------|----------------|
| 1      | Colour             | Yellowish brown|
| 2      | Odor               | Smell of Gomutra |
| 3      | Taste              | Not applicable |
| 4      | Touch              | Viscous        |
| 5      | Texture            | Liquid         |

Powder Microscopy of Intermediate product sample except arsenicals (Kalka)

Powder microscopy of Intermediate product sample (Kalka) was done with powder (60#) by studying under the Carl Zeiss Trinocular Microscope before and after staining with Phluoroglucinol and concentrated HCL to study the characters of drug. The microphotographs were taken by a camera attached with the microscope as given below.

Microscopic Characters of intermediate product sample (Kalka)

The diagnostic characters under microscope showed scleroids, spoon cells in surface view, tannin containing cells of Aragvadha fruit [Figure 9,10,11]. Presence of cluster crystal, cork cells, rosette crystal and stone cells of Dhava bark [Figure 1 to 6], Presence of cork cells, oil globules, silica deposition of Kushtha moola [Figure 7,8,20], Presence of oil globules, parenchymal cells of Haridra [Figure12,13], Presence of lignified fibres, lignified pitted vessels, stone cells of Daruharidra [Figure 14 to 19].

Pharmaceutical Analysis

Comparative Physicochemical Analysis of Sarshapa Taila and Aragvadhadi taila i.e. Refractive index, Specific gravity, Acid value, Iodine value, Saponification value were scientifically studied and results were detailed in respectively Table 5, 6.

Table 5: Comparative Physicochemical Parameters of Sarshapa Taila and Aragvadhadi taila

| Sr No. | Analytical Parameters | Result of Sarshapa Taila | Result of Aragvadhadi taila |
|--------|------------------------|--------------------------|-----------------------------|
| 1      | Refractive Index       | 1.4860                   | 1.4870                      |
| 2      | Specific gravity @ 25°C (g/ml) | 0.90                     | 0.91                        |
| 3      | Acid Value             | 5.34                     | 7.12                        |
| 4      | Iodine Value           | 8.00                     | 7.58                        |
| 5      | Saponification Value   | 288                      | 316                         |

HPTLC Study

Chromatographic study (HPTLC) was carried out under 254nm and 366nm to establish fingerprinting profile. It showed spots at 254 nm, spots at 366 nm before spray and spots at 600nm after spray.

Table 6: Results of Aragvadhadi taila

| Before/After spray | Wavelengths | No of Spots | RF value | AUC (Area Under Curve) |
|--------------------|-------------|-------------|----------|------------------------|
| Before spray       | 254 nm      | 8           | 0.03,0.07,0.11,0.14,0.20,0.28,0.69,0.97 | 13925.6, Total: 15100.4 |
|                    | 366 nm      | 5           | 0.03,0.06,0.10,0.14,0.16 | 1174.8     |
| After spray        | 600 nm      | 6           | 0.02,0.07,0.11,0.18,0.78,0.92 | -          |
DISCUSSION

Normally oils give different characteristics like colour and odor relative to ingredients which were used to prepare the medicated oil. In this Herbo mineral oil, yellowish brown colour is given due to mainly Gomutra and Aragvadh fruit pulp. The characteristic odor is due to Gomutra, Sarshapa Taila which were used in preparation. Authentication of used drugs was done by morphological and histological. This can prevent misuses of drug adulteration. The pharmacognostical evaluation shows that the intermediate product sample (Kalka) contains all ingredients which were observed in the microscopical characters. This shows the purity and quality of product.

According to present study, Saponification value of Aragvadhadi taila was 316mg/g as compared to normal value of Sarshapa Taila (288mg/g). It is the measure of average molecular weight of all fatty acids present in it. The short chain fatty acids found in fats have more saponification value. Relatively, more numbers of carboxylic functional group per unit mass of the fat. Acid values are used to measure the extent to which glycerides in the oil has been decompose by lipase and other physical factors like heat and light. Minor changes observed in acid value suggests that medicated Taila is very saturated. The iodine value is a measure of the degree of unsaturation in oil and could be used to quantify the amount of double bonds present in oil which reflects the susceptibility of oil to oxidation. This medicated oil was taken continuously 5 days of heating. However, in present study, slightly deviation was observed in Iodine value, Saponification value and Acid value as compared to normal values of Sarshapa Taila. Refractive index is an important parameter to assess quality of oil as it is change according to its compounds. Specific gravity is varying according to density of liquid which was more as compared to Sarshapa Taila. So, it suggests that the more heating gives more Saponification value, Acid value and Specific gravity. TLC fingerprint profile consists of 8.5 prominent spots under UV light at 254nm and 366nm respectively before spray and 6 were after spraying. Total Area under curve was occupy 15100.4 under UV light at 254nm and 366nm HPTLC fingerprint profile helps in identification of various phytochemical constituent present in the crude drug thereby substantiating and authenticating of product. This profile helps in identify and isolate the important phytoconstituents. These findings could be helpful in identification and authentication.

MICROPHOTOGRAPHS OF ARAGVADHADI TAILA (PLATE-1)

1. Cluster crystal of Dhava
2. Cork cells of Dhava
3. Crystal fibre of Dhava
4. Fibres of Dhava
5. Rosette crystal of Dhava
6. Stone cells of Dhava
7. Cork cells of Kusitha
8. Oil globule of Kusitha
CONCLUSION

Present study reveals that quality of *Aragvadhadi taila* as per pharmacognostical and physico-chemical parameters, which helps in justifying the quality of formulation and meet the desired quality. In the present work, the obtained results were found within normal prescribed limits. For first time, this profile of *Aragvadhadi taila* was established. On the basis of observations and experimental result, the evaluation of research of *Aragvadhadi taila* may be used as standard reference for further quality control research works and clinical studies.

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