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# Physicochemical Evaluation and Chromatographic Profiling of *Granthikadi Taila*: A Standardization Study of an Ayurvedic Medicated Oil Formulation

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## ABSTRACT

**Objective:** The present study was undertaken to establish the physicochemical characteristics and chromatographic fingerprint of *Granthikadi Taila*, a classical Ayurvedic medicated oil formulation indicated in various pediatric and neurological conditions.

**Methods:** A single sample of *Granthikadi Taila* (250 ml) was subjected to comprehensive physicochemical analysis at Cultivator Phyto Lab Private Limited, Jodhpur, following the standard protocols outlined in the Ayurvedic Pharmacopoeia of India (API). The evaluated parameters included moisture content, saponification value, acid value, iodine value, specific gravity, refractive index, rancidity test, and thin-layer chromatography (TLC) profiling.

**Results:** The analysis revealed a moisture content of 0.19% by weight. The saponification value was 160.03, acid value 5.90, and iodine value 110.78. Specific gravity at 25°C was recorded as 0.9170, and refractive index at the same temperature was 1.4712. The Kries test for rancidity was negative, indicating good oxidative stability. TLC profiling after appropriate derivatization demonstrated major spots at Rf values of 0.13, 0.18, 0.25, 0.35, and 0.68.

**Conclusion:** The analysed *Granthikadi Taila* sample exhibited physicochemical parameters characteristic of authentic medicated oil formulations. The acid value and iodine value reflect the nature of the oil base and the incorporated herbal constituents. The TLC fingerprint provides a distinctive pattern that can serve as a quality control marker for this classical Ayurvedic formulation. These findings contribute to the scientific basis for standardization of *Granthikadi Taila*.

***Keywords:*** *Granthikadi Taila*, Ayurvedic formulation, medicated oil, physicochemical analysis, standardization, thin-layer chromatography

## INTRODUCTION

Ayurvedic pharmaceutics encompasses a diverse range of dosage forms, among which *Taila* (medicated oil) preparations occupy a significant position due to their unique therapeutic applications and enhanced bioavailability of lipid-soluble active principles [1]. *Taila* formulations are prepared by processing herbal decoctions and pastes in a base oil, typically sesame oil, following classical guidelines. These preparations are extensively used in external applications like massage, internal

administration, and various therapeutic procedures such as *Nasya* (nasal instillation) and *Basti* (enema) [2].

*Granthikadi Taila* is one such classical formulation mentioned in Ayurvedic texts for the management of disorders primarily involving the nervous and musculoskeletal systems. The name "*Granthikadi*" derives from its key ingredients, which typically include *Granthika* (Piper longum) along with other herbal components. This formulation is traditionally indicated in conditions such as *Gridhrasi* (sciatica), various *Vata* disorders, and certain pediatric neurological conditions [3]. The therapeutic efficacy of such formulations depends critically on the proper manufacturing process and the quality of both the oil base and herbal ingredients.

The standardization of Ayurvedic taila preparations presents unique analytical challenges due to their complex lipid matrix and the presence of multiple heat-labile phytoconstituents [4]. Unlike modern pharmaceuticals, traditional formulations require a holistic approach to quality assessment that encompasses both classical parameters and modern analytical techniques [5]. Physicochemical parameters such as acid value, saponification value, and iodine value provide valuable information about the quality of the oil base, while chromatographic techniques offer insights into the phytochemical profile of the incorporated herbs [6].

The Ayurvedic Pharmacopoeia of India provides standardized testing protocols for various formulation types, including *Taila* preparations [7]. These parameters help in assessing the purity, stability, and overall quality of the finished product. However, published analytical data on *Granthikadi Taila* remains sparse in scientific literature. The present study was therefore designed to generate baseline physicochemical data and chromatographic profile for this important Ayurvedic formulation, which can serve as reference standards for quality control purposes.

## METHODS

### Sample Collection and Handling

A sample of *Granthikadi Taila* (250 ml) was received for analytical evaluation from Dr. Rushikesh Shivtare, PG-Department of Kaumarbhritya, Dr. Sarvepalli Radhakrishnan Rajasthan Ayurved University, Jodhpur. The sample was submitted in a plastic container and was received in unsealed condition but found to be in satisfactory state for analysis. The sample was registered on December 9, 2025, through the Test Request Form bearing the same date.

### Sample Description

Upon visual examination, the sample presented as a clear liquid with characteristic appearance consistent with medicated oil formulations. No evidence of phase separation, sedimentation, or foreign matter was observed. The sample was deemed suitable for the planned analytical procedures.

### Analytical Procedures

All analyses were performed at Cultivator Phyto Lab Private Limited, Jodhpur, during the period of December 10 to December 13, 2025. The analytical methods strictly followed the protocols described in the relevant volumes of the Ayurvedic Pharmacopoeia of India (API).

**Moisture Content:** Determined gravimetrically following API Part II Volume IV, 2017. A precisely weighed sample was heated at 105°C until constant weight was achieved. The percentage loss in weight was calculated as moisture content.

**Saponification Value:** Estimated as per API Part II Volume IV, 2017. The sample was refluxed with alcoholic potassium hydroxide solution, and the excess alkali was titrated against standard hydrochloric acid. The saponification value was expressed as milligrams of potassium hydroxide required to saponify one gram of oil.

**Acid Value:** Determined following API Part II Volume IV, 2017. The sample was

dissolved in a suitable solvent mixture and titrated against potassium hydroxide solution using phenolphthalein as indicator. The acid value represents the milligrams of potassium hydroxide required to neutralize the free acids in one gram of oil.

**Iodine Value:** Analysed as per API Part II Volume IV, 2017. This parameter measures the degree of unsaturation in oils by determining the amount of iodine absorbed by the sample under standardized conditions.

**Specific Gravity:** Measured at 25°C using a calibrated pycnometer following API Part II Volume IV, 2017. The weight of the sample was compared to the weight of an equal volume of water at the same temperature.

**Refractive Index:** Determined at 25°C using an Abbe refractometer following API Part II Volume IV, 2017. The instrument was calibrated before use, and readings were taken in triplicate.

**Rancidity Test (Kries Test):** Performed following API Part II Volume IV, 2017 to detect early stages of oxidative rancidity. The test involves the reaction of epihydrin aldehyde (if present) with phloroglucinol in the presence of mineral acids, producing a characteristic pink colour.

**Thin-Layer Chromatography (TLC):** The sample was prepared appropriately and applied on pre-coated TLC plates. The chromatogram was developed using a suitable mobile phase system. After development, the plate was derivatized with anisaldehyde sulphuric acid reagent and visualized under appropriate conditions. The retention factor (Rf) values of the major spots were calculated based on the solvent front distance of 8 cm.

### Quality Assurance

All analytical work was conducted under standard laboratory conditions with appropriate quality control measures. The laboratory maintains documented procedures, equipment calibration records, and follows good laboratory practices. The test results were reviewed and authorized by qualified scientific personnel before final reporting.

## RESULTS

### Physicochemical Parameters

The physicochemical evaluation of *Granthikadi Taila* yielded the results summarized in Table 1. The moisture content was found to be exceptionally low at 0.19% by weight, indicating minimal water content and good preservation against hydrolytic deterioration. The saponification value of 160.03 reflects the average molecular weight of fatty acids present in the oil base.

**Table 1: Physicochemical Parameters of *Granthikadi Taila***

| Parameter | Unit | Result | Test Method |
|---|---|---|---|
| Moisture | % by weight | 0.19 | API Part II Vol IV: 2017 |
| Saponification value | - | 160.03 | API Part II Vol IV: 2017 |
| Acid value | - | 5.90 | API Part II Vol IV: 2017 |
| Iodine value | - | 110.78 | API Part II Vol IV: 2017 |
| Specific gravity (at 25°C) | - | 0.9170 | API Part II Vol IV: 2017 |
| Refractive Index @25°C | - | 1.4712 | API Part II Vol IV: 2017 |
| Rancidity Test (Kries Test) | - | Absent | API Part II Vol IV: 2017 |

The acid value was recorded at 5.90, which represents the free fatty acid content of the formulation. The iodine value of 110.78 indicates a relatively high degree of unsaturation in the fatty acid composition. Specific gravity at 25°C was measured as 0.9170, and the refractive index at the same temperature was 1.4712. The Kries test for rancidity was negative, confirming the absence of oxidative deterioration in the sample.

### Thin-Layer Chromatographic Profile

TLC analysis of *Granthikadi Taila*, after derivatization with anisaldehyde sulphuric acid reagent, revealed a characteristic pattern of separated phytoconstituents. The major spots were observed at Rf values of 0.13,

0.18, 0.25, 0.35, and 0.68 (Table 2). These spots represent various phytochemicals extracted from the herbal ingredients during the manufacturing process and incorporated into the oil matrix.

**Table 2: TLC Profile of *Granthikadi Taila***

| Parameter | Details |
|---|---|
| Mobile Phase | As per API specifications |
| Derivatization | Anisaldehyde Sulphuric Acid |
| Solvent Distance Travelled | 8 cm |
| Major Rf Values | 0.13, 0.18, 0.25, 0.35, 0.68 |

The chromatographic pattern provides a characteristic fingerprint that can be used for identity verification and batch-to-batch consistency assessment of *Granthikadi Taila*.

## DISCUSSION

The standardization of traditional Ayurvedic formulations through modern analytical techniques represents a crucial bridge between classical knowledge and contemporary quality assurance requirements **[8].** The present study provides the first comprehensive physicochemical profile of *Granthikadi Taila*, offering reference data that can be utilized by manufacturers, quality control laboratories, and regulatory authorities.

The moisture content of 0.19% is notably low and highly desirable for oil-based formulations. Moisture in oils can promote hydrolytic rancidity, microbial growth, and phase separation **[9].** The observed value indicates proper manufacturing practices with adequate heating during preparation to eliminate water, as well as good storage conditions that have prevented moisture ingress. This low moisture content contributes to the extended shelf life of the formulation.

The saponification value of 160.03 provides information about the average molecular weight of triglycerides in the oil base. This value is characteristic of the specific oil used in the formulation, which is typically sesame oil in classical Ayurvedic taila preparations **[10].** Sesame oil generally has saponification values in the range of 188-195, but the slightly lower value observed in *Granthikadi Taila* may be attributed to the incorporation of lipid-soluble herbal constituents that alter the overall saponification characteristics. The saponification value is useful for detecting adulteration with other oils that have significantly different fatty acid chain lengths.

The acid value of 5.90 is relatively higher than that typically observed in fresh oils but is within acceptable limits for processed Ayurvedic formulations **[11].** Acid value represents the free fatty acid content, which can increase during processing due to heat exposure and during storage due to hydrolytic rancidity. The herbal ingredients themselves may contribute acidic compounds to the formulation. While lower acid values are generally preferable, the observed value does not indicate significant deterioration, especially considering that the rancidity test was negative.

The iodine value of 110.78 indicates a high degree of unsaturation in the fatty acid composition. This is characteristic of oils rich in oleic and linoleic acids, such as sesame oil, which is the traditional base for most Ayurvedic taila preparations **[12].** Sesame oil typically has iodine values between 103-116, and the observed value confirms that sesame oil was likely used as the base. The high unsaturation contributes to the therapeutic properties of the oil but also makes it more susceptible to oxidative rancidity. The negative Kries test suggests that oxidative deterioration has not yet occurred, indicating good manufacturing practices and appropriate storage conditions.

Specific gravity (0.9170) and refractive index (1.4712) are physical constants that provide additional identity markers for the formulation. These values are consistent with sesame oil-based preparations and serve as

rapid screening parameters for quality verification **[13].** The refractive index is particularly useful as it can be measured quickly with minimal sample preparation.

The TLC fingerprint obtained in this study is perhaps the most valuable contribution to quality control, as it provides visual evidence of the phytochemical profile of the formulation **[14].** The five major spots at Rf values 0.13, 0.18, 0.25, 0.35, and 0.68 represent compounds of varying polarities that have been successfully extracted from the herbal ingredients during the manufacturing process. Anisaldehyde sulphuric acid reagent is known to produce characteristic colours with terpenoids, steroids, and phenolic compounds **[15].** The observed pattern can serve as a reference standard for evaluating batch-to-batch consistency and for detecting adulteration or substitution.

The presence of multiple spots confirms that the manufacturing process has effectively extracted and incorporated phytoconstituents from the herbal ingredients into the oil base. The distribution of spots across the chromatogram indicates a diverse range of compounds with different polarities, which is desirable from a therapeutic perspective as it suggests the presence of multiple bioactive constituents.

While this study provides valuable baseline data, it is important to acknowledge certain limitations. The analysis was conducted on a single sample, and while the results are informative, they may not fully represent the variability that can occur across different manufacturing batches. Factors such as variations in raw material quality, manufacturing procedures, and storage conditions can influence the final product's characteristics **[16].** Additionally, the study did not attempt to identify or quantify specific marker compounds from individual herbal ingredients.

Future research should focus on analysing multiple batches from different manufacturers to establish acceptable ranges for each parameter. Development and validation of high-performance thin-layer chromatography (HPTLC) methods would enable more precise quantification of marker compounds. Correlation of physicochemical parameters with therapeutic efficacy through controlled clinical studies would provide valuable insights into the relationship between quality attributes and clinical outcomes **[17].**

The findings of this study have practical implications for the Ayurvedic pharmaceutical industry. Manufacturers can use these parameters as in-process and finished product quality indicators. Regulatory authorities can consider these values when developing monographs for *Granthikadi Taila* in official pharmacopoeias. Practitioners can have greater confidence in the quality of formulations that meet these analytical specifications.

## CONCLUSION

The physicochemical characterization of *Granthikadi Taila* has established baseline parameters that can serve as quality reference standards for this classical Ayurvedic formulation. The low moisture content, characteristic saponification and iodine values, and negative rancidity test indicate a well-formulated and properly stored product. The TLC fingerprint with major spots at Rf 0.13, 0.18, 0.25, 0.35, and 0.68 provides a distinctive pattern for identity verification. These findings contribute to the scientific evidence base for *Granthikadi Taila* and support its quality control and standardization. Further studies involving multiple batches and marker compound quantification would enhance the quality assurance framework for this important Ayurvedic formulation.

### *Declaration by Authors*

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