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Pharmaceutical Standardization and HPTLC Fingerprinting of Erandamooladi Fume Stick (Dhoomavarti): A Herbal Formulation

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Abstract

Introduction: Ensuring the identity, quality, safety, and reproducibility, standardizing a drug has become a cornerstone of the drug in the modern era. When treating illnesses in the Supraclavicular *(Urdhwajatrugata)* region, a special type of medicated fume inhalation therapy (Dhoomanasya) using medicated fume sticks *(Dhoomavarti)* is mentioned in Ayurveda. The HPTLC fingerprinting technique is a commonly used approach for herbal formulation quality monitoring and hence can be used to monitor the quality of the medicated fume stick.
Aim: To evaluate the pharmaceutical and analytical profile of Erandamooladi fume stick *(Dhoomavarti)* through organoleptic evaluation, physicochemical parameters, and HPTLC fingerprinting.
Materials and Methods: The formulation was prepared in a single batch using authenticated raw drugs: Erandamoola (*Ricinus communis*), Jatamansi (*Nardostachys jatamansi*), Guggulu (*Commiphora mukul*), Aguru (*Aquilaria agallocha*), and Chandana (*Santalum album*). Organoleptic and physicochemical parameters like loss on drying, ash value, extractive values, carbon content, and volatile oil content and some specific phytochemical tests were performed as per standard guidelines. HPTLC fingerprinting was performed using methanol extract of the formulation and the developed plate was observed under UV light.
Results: The formulation was brown in colour, hard in consistency, and had a camphor-like odour. Loss on drying was 5.8%, total ash value was 5%, and acid insoluble ash was found to be 2.3%. Water-soluble extract was 14% and alcohol-soluble extractive was 18%. Carbon content was 0.3% per varti and volatile oil content was 0.4%. HPTLC profile showed multiple bands under UV light, indicating the presence of diverse phytoconstituents.
Conclusion: These analytical findings provides baseline standardization parameters and establishes the HPTLC fingerprint of this herbal formulation. These findings may be useful for future quality control, reproducibility, and research validation.

INTRODUCTION

Due to the need of the modern scientific world, Ayurveda has begun to place a strong importance on the need of well-prepared medications in order to achieve therapeutic efficacy. The identity and purity of the basic ingredients as well as the preparation, stability, and storage methods all affect the quality of an Ayurvedic formulation. To increase the acceptability and reproducibility of Ayurvedic medicines in scientific research and international health systems, standardization and analytical evaluation have become crucial in the modern day.[1]

Medicated Dume Stick *(Dhoomavarti)* is a special dose form used for Medicated Fume Inhalation (Dhoomapana or Dhooma Nasya), which entails the oral and nasal administration of therapeutic fumes. While Medicated fume inhalation through nose (Dhooma Nasya) is mainly acts on Supraclavicular region (Urdhvajatrugata regions) and acts on conditions of head and neck region) like different types of headaches (shirahshoola), eye disorders (Netra roga), Ear disorders (Karna roga) and others, Medicated fume inhalation through mouth *(Dhoomapana)* is believed to mainly act on the respiratory system *(Pranavaha Srtotas)* of the patient. Thus the therapeutic benefits of medicated fume inhalation are splendidly discussed in relation to illnesses affecting the respiratory system and suplaclavicular region including head. [2,3]

Erandamooladi fume stick *(Dhoomavarti)* is a polyherbal formulation mentioned in Charaka Samhita which contains Erandamoola (Ricinus communis), Jatamansi (Nordostachys jatamnsi), Guggulu (Commiphora mukul), Aguru (Aquillaria agallocha), and Chandana (Santalum album). These drugs are traditionally considered to possess analgesic (Shoolahara), anti-inflammatory and pain stabilizing *(Vedanasthapana)* properties, and is mentioned in the treatment of headaches (Shirahshoola). [4]

The most common modern analytical tool used for herbal fingerprinting and standardization these days is High Performance Thin Layer Chromatography (HPTLC). It is mainly useful in analysis of complex polyherbal formulations where multiple phytoconstituents contribute to therapeutic action.[5] Establishing an HPTLC fingerprint profile helps in quality assurance and provides a reproducible reference for future batches.[6]

Therefore, the present work was undertaken to prepare Erandamooladi

Fume stick *(Dhoomavarti)* and evaluate its pharmaceutical parameters and HPTLC fingerprint profile.

AIM AND OBJECTIVES

Aim

To establish the pharmaceutical and analytical standardization parameters of Erandamooladi Fume stick*(Dhoomavarti)*.

Objectives

1. To authenticate the raw drugs used in the preparation of Erandamooladi Fume stick*(Dhoomavarti)*.
2. To prepare Erandamooladi Fume stick*(Dhoomavarti)* using a standard operating method.
3. To evaluate the formulation through organoleptic and physicochemical parameters.
4. To develop an HPTLC fingerprint profile using methanolic extract.

MATERIALS AND METHODS

Collection and Authentication of Raw Drugs

The raw drugs were procured from Vadodara market, Gujarat, India. Botanical authentication was carried out by the Department of Dravyaguna, Parul Institute of Ayurveda and Research.

The authenticated botanical names and plant parts are shown in Table 1.

Table 1: Authentication of Raw Drugs

Sr. No.	Common Name	Botanical Name	Part Used
1	Eranda (Castor)	*Ricinus communis*	Root
2	Jatamansi (Spikenard)	*Nardostachys jatamansi*	Root
3	Guggulu (Indian Myrrh)	*Commiphora mukul*	Resin
4	Aguru (Agarwood)	*Aquilaria agallocha*	Wood
5	Chandana (Sandal wood)	*Santalum album*	Wood

Method of Preparation of Fume stick*(Dhoomavarti)*

The preparation method was adopted with reference to the published procedure of Fume stick*(Dhoomavarti)* preparation.[7]

The steps followed were:

- A 24 cm long reed (Ikshika) was taken and soaked overnight in water.
- A cotton cloth of size 16 × 16 cm was washed in hot water and dried.
- Kalka (paste) weighing 12 g was prepared and uniformly applied on the cloth.
- A piece reed was placed at one end of the cloth.
- The cloth was rolled into approximately five layers.
- The rolled material was shade dried completely.

- Finally, a Fume stick*(Dhoomavarti)* weighing approximately 12 g was obtained.

The formulation was prepared in a single batch for the purpose of clinical study.

Organoleptic Evaluation

The prepared Fume stick*(Dhoomavarti)* was evaluated for colour, odour, consistency, and appearance by sensory evaluation. Organoleptic evaluation is considered a primary step in herbal drug standardization.[8]

Physicochemical Evaluation

To evaluate the quality of the prepared formulation, Physicochemical analysis was performed. The Ayurvedic Pharmacopoeia of India's conventional pharmacopeial processes were followed in evaluating parameters such as extractive values, ash values, and loss on drying.[1]

The parameters studied were:

- Loss on drying at 110°C
- Total ash value
- Acid insoluble ash
- Water soluble extractive value
- Alcohol soluble extractive value
- Carbon content
- Volatile oil content

Phytochemical Analysis

Other specific preliminary phytochemical screening were also carried out to identify the presence of major phytoconstituents. The analysis was performed using standard qualitative chemical tests to detect alkaloids, flavonoids, tannins, phenolic compounds, and saponins. The test sample was prepared by dissolving a small quantity of powdered Fume stick*(Dhoomavarti)* in methanol, followed by filtration. The filtrate was used for further analysis. [9]

Hager's test (Alkaloids)

Few drops of Hager's reagent (saturated picric acid solution) were added to 2ml of the test solution.

Shinoda test (Flavonoids)

A small quantity of magnesium ribbon was added to 2 ml test solution followed by few drops of concentrated hydrochloric acid.

Ferric chloride test (Phenols and tannins)

2–3 drops of ferric chloride solution were added to 2ml test solution.

Foam test (Saponins)

About 5 mL of the test solution was shaken vigorously in a test tube for 2 minutes and allowed to stand.

Salkowski test (Steroids and triterpenoids)

2 mL of chloroform was added to 2 ml of test solution followed by careful addition of concentrated sulphuric acid along the sides of the test tube.

HPTLC Fingerprinting

For the HPTLC fingerprinting of the Erandamuladi Fume Stick (Dhooma Varti), Methanolic extract of the formulation was used as it is the widely used component for extraction of a broad range of

phytoconstituents including alkaloids, flavonoids, terpenoids and phenolic compounds.[10][11] Using the CAMAG HPTLC system and the vision CATS software (Server HPTLC v3.2.23095.1), the analysis was performed. The Linomat 5 applicator was used to apply the sample, and the TLC Scanner 4 was used for scanning. Chromatographic development took place in a twin trough chamber measuring 10 × 10 cm.

The stationary phase was Merck HPTLC silica gel 60 F_{254} plates (100 × 100 mm). The samples had a band length of 8.0 mm and were applied at a Y-position of 8.0 mm. There was a constant 22.4 mm between each track. Up to an 80 mm solvent front distance, the plate was developed.

Chloroform : Methanol : Formic acid in the ratio of 7 : 3 : 0.2 v/v/v was used as a mobile phase. The formulation extract was applied in four different volumes (which are 5 µL, 10 µL, 15 µL and 20 µL) in order to obtain a clear and comparative chromatographic pattern. The plate was then scanned at 254 nm (absorbance mode), 366 nm (fluorescence mode), and 210 nm (absorbance mode). Rf values of the resolved spots were recorded and used to establish the fingerprint profile.

OBSERVATIONS AND RESULTS

Organoleptic Characters

The organoleptic evaluation showed the following characters:

- Colour: Brown
- Odour: Camphor-like
- Taste: Not assessed
- Consistency: Hard brown solid

Physicochemical Parameters

The physicochemical evaluation results are as shown in Table 2.

Table 2: Physicochemical Parameters of Erandamooladi Fume stick*(Dhoomavarti)*

Parameter	Result
Loss on drying at 110°C	5.8%
Total ash value	5%
Acid insoluble ash	2.3%
Water soluble extractive	14%
Alcohol soluble extractive	18%
Carbon content	0.3% per varti
Volatile oil content	0.4%

Phytochemical Tests

The qualitative phytochemical analysis of the methanolic extract of Fume stick*(Dhoomavarti)* showed the presence of important bioactive phytoconstituents. These constituents may contribute to the pharmacological potential of the formulation.

Phytoconstituent group	Test performed	Observation	Result
Alkaloids	Hager's test	Yellow precipitate	Present (+)
Flavonoids	Shinoda test	Pink coloration	Present (+)
Phenols and tannins	Ferric chloride test	Greenish-black colour	Present (+)
Saponins	Foam test	Persistent froth	Present (+)
Steroids / triterpenoids	Salkowski test	Reddish-brown ring	Present (+)

HPTLC Fingerprinting

HPTLC analysis of the methanolic extract of Erandamuladi Fume stick*(Dhoomavarti)* presented a distinct chromatographic fingerprint pattern at different wavelengths. At 254 nm (Table-3), the spots were not prominent in the 5 µL track. However, a major spot was clearly observed at Rf 0.938 in both 10 µL and 15 µL tracks. In the 20 µL track, the major spot was observed at Rf 0.921, suggesting a similar compound profile with slight variation in peak prominence due to higher concentration.

At 366 nm (Table-4), multiple fluorescent spots were detected, indicating the presence of several phytoconstituents. The Rf values ranged from 0.071 to 0.936, and the number of spots increased with higher sample application volume. This suggests that lower volume application may not be sufficient to visualize all phytochemical constituents of the formulation.

At 210 nm (Table-5), the chromatogram showed additional spots, particularly in the 10 µL, 15 µL and 20 µL tracks. No spots were detected in the 5 µL track at this wavelength. The detected Rf values ranged from 0.235 to 0.978, supporting the presence of UV-absorbing compounds such as aromatic and resinous phytoconstituents.

The observed Rf values at 254 nm, 366 nm and 210 nm are presented in Tables 3–5. The developed plate photograph under UV light is shown in Figure 1. The overall HPTLC profile obtained may serve as a preliminary fingerprint standard for Erandamuladi Fume stick*(Dhoomavarti)*.

Table 3: Rf values of Erandamuladi Fume stick*(Dhoomavarti)* at 254 nm

Track No.	Volume (µL)	Major Rf Value
1	5.0	Not prominent

Track No.	Volume (µL)	Major Rf Value
2	**10.0**	**0.938**
3	**15.0**	**0.938**
4	**20.0**	**0.921**

Table 4: Rf values of Erandamuladi Fume stick*(Dhoomavarti)* at 366 nm

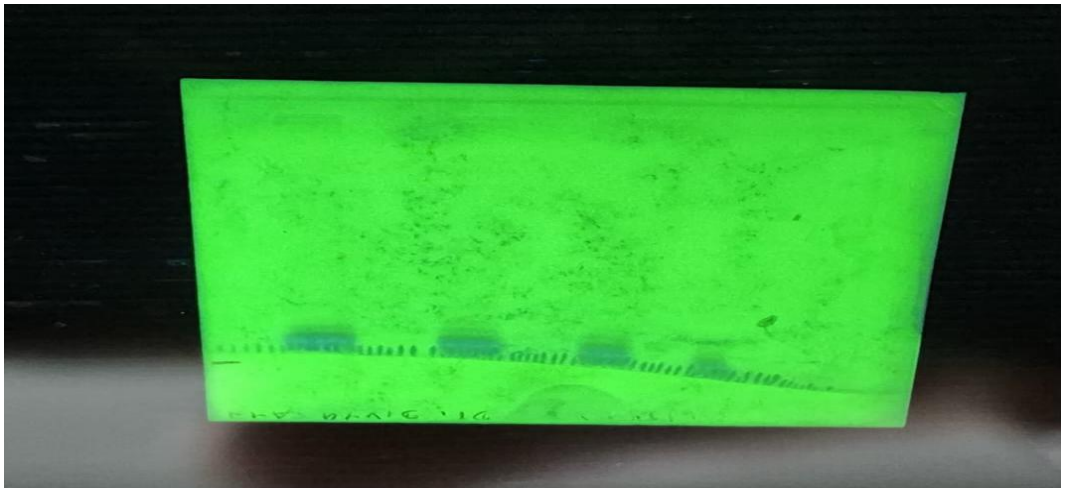
Track No.	Volume (µL)	Observed Rf Values
1	5.0	0.081
2	10.0	0.085, 0.936
3	15.0	0.074, 0.326, 0.728
4	20.0	0.071, 0.331, 0.829

Table 5: Rf values of Erandamuladi Fume stick*(Dhoomavarti)* at 210 nm

Track No.	Volume (µL)	Observed Rf Values
1	5.0	Not detected
2	10.0	0.247, 0.769, 0.978
3	15.0	0.237, 0.714
4	20.0	0.235, 0.568, 0.718

The presence of several bands indicates a diverse phytochemical composition, supporting the polyherbal nature of the formulation.[12] This fingerprint profile can be used as a standard reference for future quality control and standardization studies.

.**Figure 1: HPTLC Plate Photograph of Erandamooladi Fume stick*(Dhoomavarti)* under UV Light**



DISCUSSION

Standardization of Ayurvedic formulations plays a vital role in ensuring reproducibility and credibility of clinical research.[1] In this study, Erandamooladi Fume stick *(Dhoomavarti)* showed the organoleptic characters such as brown colour and camphor-like odour were consistent with the aromatic nature of ingredients like Agar wood *(Aguru- Aquilleria agallocha)*,Sandalwood *(Chandana- Santalum album)* and *Spikenard (Jatamansi- Nordostachys jatamansi)*. Organoleptic evaluation is a basic yet reliable preliminary tool for assessing herbal formulations.

Loss on drying (5.8%) indicates low moisture content, suggesting that the formulation was properly dried. Control of moisture content is essential to prevent microbial contamination and enhance shelf stability of herbal preparations. Total ash value (5%) reflects the total inorganic residue present in the formulation.[1] Acid insoluble ash (2.3%) represents silica content and helps estimate contamination with sand or earthy materials. [1] In the present formulation, ash values suggest minimal contamination and acceptable purity. Extractive values are useful indicators of the solubility of active constituents. [1] Water soluble extractive value (14%) shows the presence of polar phytoconstituents. Alcohol soluble extractive value (18%) was comparatively higher, which may be attributed to resinous and aromatic components present in ingredients such as Indian Myrrh *(Guggulu- Commiphora mukul)* , Agar wood *(Aguru- Aquillaria agallocha)* and Sandal wood *(Chandana- Santalum album)*. Such constituents may contribute to the therapeutic effect of the formulation. Volatile oil content (0.4%) indicates the presence of aromatic essential oils, which are particularly relevant in Fume stick*(Dhoomavarti)* formulations as

volatile components are likely to be delivered through medicinal fumes. [5] Essential oils are acknowledged to have anti-inflammatory and analgesic properties[5], which provides the justification for the traditional use of Fume stick*(Dhoomavarti)* in the pain management of the supraclavicular region *(urdhwajatrugata sthana)*. The preliminary phytochemical analysis indicated the presence of alkaloids, flavonoids, tannins, phenolic compounds, saponins, and steroids/triterpenoids in the formulation. These phytoconstituents are well known for their anti-inflammatory, analgesic, antioxidant, and neuroprotective potential. Hence, the presence of these bioactive constituents may provide a scientific basis for the traditional use of Fume stick*(Dhoomavarti)* in managing pain-related conditions of the supraclavicular region *(urdhwajatrugata region)*.[13]

Further, HPTLC fingerprinting showed characteristic chromatographic pattern at 254 nm, 366 nm and 210 nm, indicating the presence of diverse chemical constituents. At 254 nm, the presence of a prominent spot around Rf 0.938 in 10 µL and 15 µL tracks indicates the presence of a major UV-absorbing compound in the formulation. The 20 µL track also showed a major spot near Rf 0.921, which may represent the same compound with a slight shift due to concentration variation and band diffusion. The 5 µL track did not show prominent spots, suggesting that lower sample loading may not be sufficient to detect key constituents at this wavelength. At 366 nm, the chromatogram displayed multiple fluorescent spots with Rf values ranging between 0.071 and 0.936, suggesting the presence of compounds such as volatile aromatic constituents, resinous fractions, and plant secondary metabolites.[5] This observation is relevant because Erandamuladi fume stick*(Dhoomavarti)* contains ingredients like Agarwood *(Aguru- Aquillaria agallocha)*, Sandalwood *(Chandana- Satalum album)*, Spikenard *(Jatamansi- Nordostachys jatamansi)* and Indian Myrrh *(Guggulu- Commiphora mukul)*, which are known for aromatic and resinous chemical profiles. These components may contribute to the therapeutic utility of the formulation when used for therapeutic inhalation through mouth (Dhoomanasya), as volatile constituents are expected to play an important role during inhalational administration. At 210 nm, additional spots were detected with Rf values extending up to **0**.978, which indicates the presence of compounds with strong UV absorption. The increased number of spots at higher sample application volumes supports the importance of adequate concentration during analysis for better visualization of phytoconstituents.[12] Overall, the observed

Rf values and the presence of multiple bands under UV light indicates a complex phytochemical composition. HPTLC fingerprinting provides a chemical identity of the formulation and is widely accepted as a quality control tool for herbal drugs. Hence, this study can be used as a reference standard for ensuring batch-to-batch consistency, quality control and detection of adulteration thus supporting the future pharmaceutical studies in this area.

CONCLUSION

The present study establishes pharmaceutical standardization parameters of Erandamooladi Fume stick *(Dhoomavarti)* prepared in a single batch. HPTLC fingerprinting at 254 nm, 366 nm and 210 nm revealed reproducible spots with Rf values extending from 0.071 to 0.978, suggesting the presence of multiple phytoconstituents. The established chromatographic profile may be used as a baseline standard for future quality control of Erandamuladi Fume stick *(Dhoomavarti)*.

LIMITATIONS

The study was limited to a single batch preparation. Marker-based HPTLC quantification and densitometric peak analysis were not performed due to non-availability of reference standards.

FUTURE SCOPE

Further evaluation including microbial load, heavy metal analysis, pesticide residue testing, and densitometric HPTLC profiling using marker compounds may improve global acceptance of this formulation.

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CONFLICT OF INTEREST

There is no conflict of interest.

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None.

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