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A PHARMACEUTICO-PHARMACOGNOSTICAL AND CHROMATO GRAPHIC EVALUATION OF EDAGAJADI LEPA: A NOVEL AYURVEDIC ANTI-FUNGAL APPROACH

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ABSTRACT

Edagajadi Lepa (Ayurvedic antifungal formulation) is a potent compound mentioned by Acharya Charaka, Acharya Sushruta and Acharya Vagbhata as a part of treatment of Skin disease. It treats most of skin condition such as Fungal infection, Psoriasis, Helminthic etc., diseases. Edagajadi Lepa is a polyherbal formulation which comprises of five herbal drugs and one fermented preparation. This all ingredients are having Anti-pruritic, Anti-bacterial, Anti-oxidant, Anti-inflammatory and Anti-helminthic activities. Because of its wide therapeutic potential, Edagajadi Lepa serves as a unique bridge between traditional wisdom and modern pharmaceutics. The aim of study was to prepare Edagajadi Lepa and authenticate by physico-chemical and Pharmacognostic parameters. Pharmacognostic, Pharmaceutical TLC studies were done at pharmaceutical chemistry and Pharmacognosy department of ITRA, Jamnagar. While, HPTLC study was carried out at Vasu research centre, Vadodara, Gujarat. Pharmacognostic findings for Edagajadi Lepa was used for confirm the authenticity and genuine ingredients of this Ayurvedic formulation. The average values of physicochemical parameters of Edagajadi Lepa were found as follows: pH value: 6, loss on drying: 6.04%, Total Ash value: 18.68%, Water soluble extractive value: 59.30%, Methanol soluble extractive value:

40.82%, Petroleum ether: 20.17%, Qualitatively TLC study showed seven spots at 254 nm and twelve spots at 366 nm. HPTLC study shows prominent peaks (five at 254 nm, six at 366 nm and eight at 540 nm). This results of Edagajadi Lepa will be used to develop a preliminary standard profile for this Antifungal drug. The data generate from this described the purity, quality and efficacy of Ayurvedic formulation.

Keywords: Edagajadi Lepa, Poly-herbal formulation, Anti-fungal, Pharmacognostical, Pharmaceutical, HPTLC fingerprinting

INTRODUCTION

Ayurveda is ancient Indian system of medicine; it not only emphasizes the treatment of diseases but also focuses on their prevention and promote the healthy lifestyle. Many formulations giving in Ayurvedic classical texts for the Kustha Roga (skin disease) among them one is Edagajadi Lepa which is mentioned in Charaka Samhita under the context of Dadru Kustha (fungal infection) by Acharya Charaka **[1]**. It contains *Cassia tora* (Edagaja), *Saussurea lappa,* (Kustha), *Rock salt* (Saindhava Lavana), *Embelia ribes* (Vidanga), *Brassica compestris* (Shweta Sarshapa) and fermented preparation (Sauveeraka).

This Edagajadi Lepa having Katu-Tikta Rasa (pungent and bitter taste), Laghu-Ruksha Guna (light and dry qualities), Ushna-Tikshna Veerya (hot and penetrating potency), Kandughna (anti-pruritic), Kushthaghna (anti-dermatotic) and Krimighna (anti-microbial) properties **[2]**. Edagajadi Lepa is primarily indicated for external application in various Kushtha Rogas (skin disorders) and Kandu (itching) conditions. The formulation acts by pacifying Kapha and Pitta Dosha (body humors), removing Krimi (microbial or parasitic infestations), Kandu (itching) and Sotha (inflammation). The synergistic action of its ingredients helps in detoxifying the skin and restoring normal physiological balance.

In the current scenario of the market, where the purity and quality of herbal drugs are easily compromised due to improper identification, adulteration and by environmental factors, it becomes essential to authenticate and standardize such classical formulations and it is also important to reassure the herbal drugs before bringing them to market. Standardization requires for each and every formulation because it ensures the safety, efficacy and reproducibility of Ayurvedic medicine. Different types of quality control tools are used to assure the quality of the drugs. TLC study is used for qualitative and semi-quantitative analysis of natural products for separation and analysis of the mixture of compounds **[3]**. HPTLC study is useful for

the qualitative and quantitative analysis, and the detection of adulterants or any impurities **[4]**. Hence, this study is aimed at authentication and development of analytical profile of Edagajadi Lepa through modern scientific parameters, ensuring its identity, purity and consistency for safe and therapeutic use.

Aim

- The study is aimed to prepare Edagajadi Lepa (Ayurvedic antifungal formulation) and authenticate it through physico-chemical evaluation.

Objectives

1. To identify and authentication of the raw materials used in the preparation and standardize the preparation process of Edagajadi Lepa.
2. To conducting Pharmaceutico-analytical studies to evaluate the organoleptic, physicochemical, and qualitative parameters of the Edagajadi Lepa.

MATERIALS AND METHODS

The pharmaceutical study of Edagajadi Lepa (Ayurvedic antifungal formulation) was carried out at Pharmacy. ITRA, Jamnagar, Gujarat, through the following steps:

1. Collection of raw drugs for Edagajadi Lepa
2. Preparation of Edagajadi Lepa Powder
3. Preparation of Fermented Preparation (Sauveeraka)

1. Collection of raw drugs for Edagajadi Lepa

Most of the raw drugs of Edagajadi Lepa were collected from the Pharmacy, ITRA, Jamnagar. Shweta Sarshapa was collected from the Pharmacy of Surat, Gujarat. All ingredients are mentioned below in **(Table 1)**.

2. Preparation of Edagajadi Lepa powder

All ingredients such as, Edagaja Beeja, Kushtha, Saindhava, Shweta Sarshapa and Vidanga are taken in equal quantity and make powdered, mixed well and stored in air-tight containers **(Figure 1)**.

Table 1: Formulation compositions of Edagajadi Lepa [5]

SN	Ingredients	Latin/ English name	Part used [6]	Proportion
1.	Edagaja	*Cassia tora* Linn.	Seed	1 part
2.	Kushtha	*Saussurea lappa,* C.B. Clarke	Root	1 part
3.	Saindhava	*Rock salt*	Powder	1 part
4.	Shweta Sarshapa	*Brassica compestris* Linn.	Seed	1 part
5.	Vidanga	*Embelia ribes* Burm. F	Fruit	1 part
6.	Sauveeraka	Fermented preparation	-	Q.S.

Fig 1(a): All ingredients are taken in equal proportion

Fig 1(b): Mixed well in Edge Runner

Fig 1(c): Dried and stored in air tight container

Fig 1(d): Edagajadi Lepa application

Figure 1: Steps of Edagajadi Lepa Powder Preparation.

3. Preparation of Fermented Preparation (Sauveeraka)

For Sauveeraka the preparation, *Hordeum vulgare* (Yava) was de-husked and ground in a Khalva Yantra (mortar and pestle). After that, eight times water was added and boil until it reduced to one half. The preparation was then transferred to an earthen pot and kept undisturbed for 15 days. after the completion of fermentation process, the lid was opened and filtration was carried out **[7]**. *Sauveeraka* was prepared in the Agad Tantra Department of I.T.R.A. Jamnagar **(Figure 2)**.

Fig 2(a): *H. vulgare* (Yava) de-husked in mortar and pestle

Fig 2(b): *H. vulgare* (Yava) mixed with 8 times water and boil till reduce to half

Fig 2(c): Take in earthen pot

Fig 2(d): Close lid and kept for 15 days (undisturbed)

Figure 2: Steps of Sauveeraka Preparation

Pharmacognostic evaluation:

The drugs which were used in this formulation of Edagajadi Lepa was identified and authenticated by the Pharmacognosy Laboratory, ITRA, Jamnagar **[8]**.

Powder microscopy

All the Powdered drugs were studied microscopically and microscopic characters of individual drugs were noted. The powder of the whole drug was dissolved with water followed by microscopy of the sample without stain and after staining with Phloroglucinol + HCl. Microphotographs of the sample were also taken under Carl – Zeiss trinocular microscope **[9]**.

Table 2: Microscopical characters of Edagajadi Lepa

No.	Drug	Microscopic characters
1	*Cassia tora* Linn (Edagaja)	Endocarp cells, Lignified fibres and Stone cell with Brown content of Edagaja
2	*Saussurea lappa,* C.B. Clarke (Kustha)	Fibre of Kushtha, Oleoresin with crystals of Kushtha, Pitted vessels of Kushtha
3	*Brassica compestris* Linn. (Shweta Sarshapa)	Epicarp cells of Sarshapa, Fibres of Sarshapa, Oil globules of Sarshapa
4	*Embelia ribes* Burm. F (Vidanga)	Resin content of Vidanga, Stone cell of Vidanga

Pharmaceutical Analysis of Edagajadi Lepa:

Pharmaceutico-analytical study was carried out at pharmaceutical laboratory, ITRA, Jamnagar.

Qualitative Analysis consideration:

Physico-chemical analysis was conducted to find out the following parameters,

- pH value
- Loss on drying
- Total Ash value
- Water Soluble Extract
- Methanol soluble Extract
- Petroleum ether (%)

Thin Layer Chromatography (TLC):

Thin layer Chromatography (TLC) study was conducted at pharmaceutical laboratory, ITRA, Jamnagar.

Materials & Methods Section

The TLC method is at present an important analytical tool for qualitative and semi quantitative analysis of a number of natural products. The adsorbent, such as Silica Gel G, is coated to a thickness at 0.3 mm or clean TLC plates using commercial spreader, the plates are activated at 105^0 C for 30 minutes and used. The selection of the mobile phase depends upon the type of constituents to be analysed. After the development of chromatogram by ascending technique, the resolved spots are revealed by spraying with suitable detecting agents **[10]**.

TLC conditions

Sample preparation: The Drug was powdered and was extracted with Methanol for 1 hour and then filter and filtrate was used for TLC.

Stationary Phase: Merck pre-coated silica gel 60 F254 plate

Solvent system: Toluene: Ethyl Acetate: Formic acid: Methanol (6: 3: 0.1: 1)

Solvent front: 8.5 cm

Spray reagent: Anisaldehyde Sulphuric Acid Spray

High performance thin layer chromatography (HPTLC):

High performance thin layer chromatography (HPTLC) study was carried out at Vasu Research Centre, Vadodara, Gujarat.

Sample Preparation for HPTLC

1 g of sample was weighed accurately in a conical flask. To it 10 mL methanol was added, reflux for 30 minutes on water bath. Then, allowed to cool and filtered with the help of Whatman filter paper No. 1. The

filtrate thus obtained was used for HPTLC fingerprinting **[11]**.

Preparation of Spray reagent (Anisaldehyde - sulphuric acid reagent)

0.5 mL Anisaldehyde is mixed with 10 mL Glacial acetic acid, followed by 85 mL Methanol and 5 mL Sulphuric acid (98 %).

Chromatographic Conditions

Table 3: Method for developing Chromatographic condition [12]

Application Mode	**CAMAG Linomat 5 – Applicator**
Filtering System:	**Whatman filter paper No. 1**
Stationary Phase:	**MERCK-TLC/HPTLC Silica gel 60 F254 on Aluminum sheets**
Application (Y axis) Start Position:	**10 mm**
Development End Position:	**80 mm from plate base**
Sample Application Volume:	**8.0 µL**
Development Mode:	**CAMAG TLC Twin Trough Chamber**
Chamber Saturation Time:	**30 minutes**
Mobile Phase (MP):	**Toluene: Ethyl acetate: Acetic acid (7: 2: 1 v/v)**
Visualization:	**@254nm, @366nm and @540nm (after derivatization)**
Spray reagent:	**Anisaldehyde - sulphuric acid reagent**
Derivatization mode:	**CAMAG – Dip tank for about 1 minute**
Drying Mode, Temp. & Time:	**TLC Plate Heater Preheated at 100± 5^{0}C for 3 minutes**

RESULTS:

After the preparation of Edagajadi Lepa, the observation and results were found are mentioned as below.

Finished product microscopy

Microscopic evaluation of finished product (Edagajadi Lepa) was conducted, characters were noted down and micro photographs were taken which were observed in photo microscopy in 10 X lens which is mentioned in **(Figure 3)**.

Organoleptic Parameters of Prepared Drug

It refers to analytical methods like Sparsha (consistency), Roopa (colour), Rasa (taste), Gandha (odour) etc., **[13]** One pinch of Edagajadi Lepa Churna was randomly selected and the identification was carried out based on the organoleptic features and microscopy of the prepared drug**.** Which is mentioned below in **(Table 4)**.

Physico-Chemical Analysis:

After the preparation of Edagajadi Lepa, the Physico-chemical analysis **[14]** was carried out for all three batches on different parameters and results are presented in **(Table 5)**.

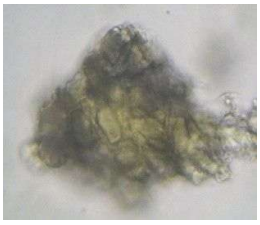			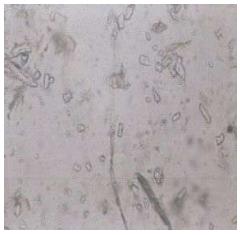
Fig 3(a): Endocarp cells of Edagaja	**Fig 3(b): Lignified fibres of Edagaja**	**Fig 3(c): Stone cell with Brown content of Edagaja**	**Fig 3(d): Fibre of Kushtha**
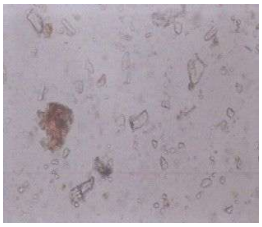	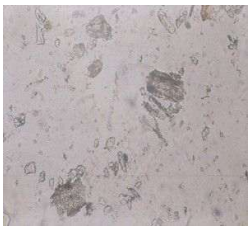		
Fig 3(e): Oleoresin crystals of Kushtha	**Fig 3(f): Pitted vessels of Kushtha**	**Fig 3(g): Epicarp cells of Sarshapa**	**Fig 3(h): Fibres of Sarshapa**
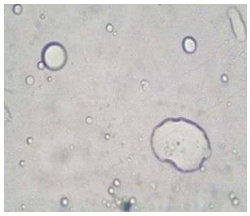	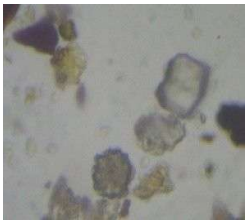	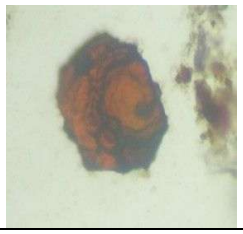	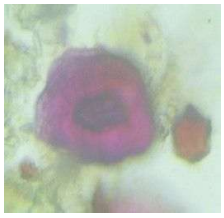
Fig 3(i): Oil globules of Sarshapa	**Fig 3(j): Calcium oxalate crystals of Vidanga**	**Fig 3(k): Resin content of Vidanga**	**Fig 3(l): Stone cell of Vidanga**

Figure 3: Microphotographs of Prepared Drug

Table 4: Organoleptic characters of Edagajadi Lepa

Characters	Results
Sparsha (consistency)	Frictionless
Rupa (colour)	Dark brownish
Rasa (taste)	Characteristic
Gandha (odour)	Characteristic

Table 5: Physico-Chemical parameters of Edagajadi Lepa

Analytical Parameters	Edagajadi Lepa
pH Value	6
Loss on drying (%)	6.04%
Total Ash value (%)	18.68 %
Water soluble extract (% w/w)	59.30 %
Methanol soluble extract (%)	40.82 %
Petroleum ether (%w/w)	20.17 %

TLC value of Edagajadi Lepa:

TLC fingerprinting was one of the fundamental objectives of present study. Below are the images of Edagajadi Lepa TLC showing the separation of components at different levels, at different wavelengths and after spray. TLC showed 7 spots under 254 nm, 12 spots under 366 nm and 6 spots after spraying with Anisaldehyde sulphuric acid spray. TLC study with Rf values of all spots and their colours are mentioned in **(Table 6)** and photographs **(Figure 4)**.

Table 6: TLC Rf values of Edagajadi Lepa

VU light	Spot found at different Rf Value
UV 254 nm	0.18, 0.36, 0.40, 0.48, 0.63, 0.75, 0.88
UV 366 nm	0.05, 0.18, 0.21, 0.27, 0.31, 0.35, 0.46, 0.56, 0.63, 0.79, 0.82, 0.89
After derivatized with Anisaldehyde Sulphuric acid Spray	0.18, 0.29,0.37, 0.50, 0.75, 0.87

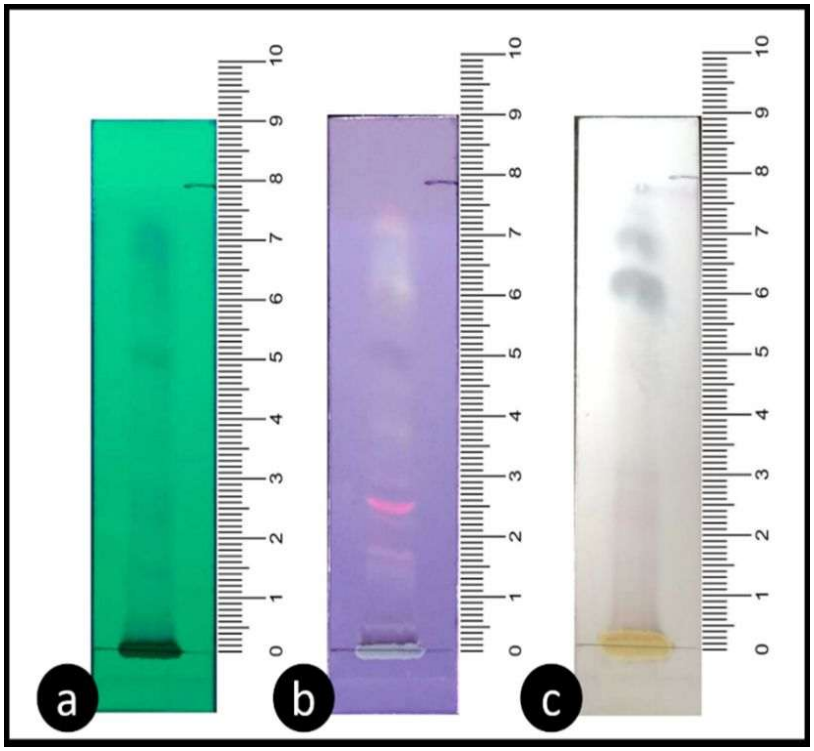

Figure 4: Photographs of TLC study of Edagajadi Lepa (*a*- UV 254 nm, b- UV 366 nm, c- After derivatized with Anisaldehyde Sulphuric Acid Spray)

HPTLC value of Edagajadi Lepa

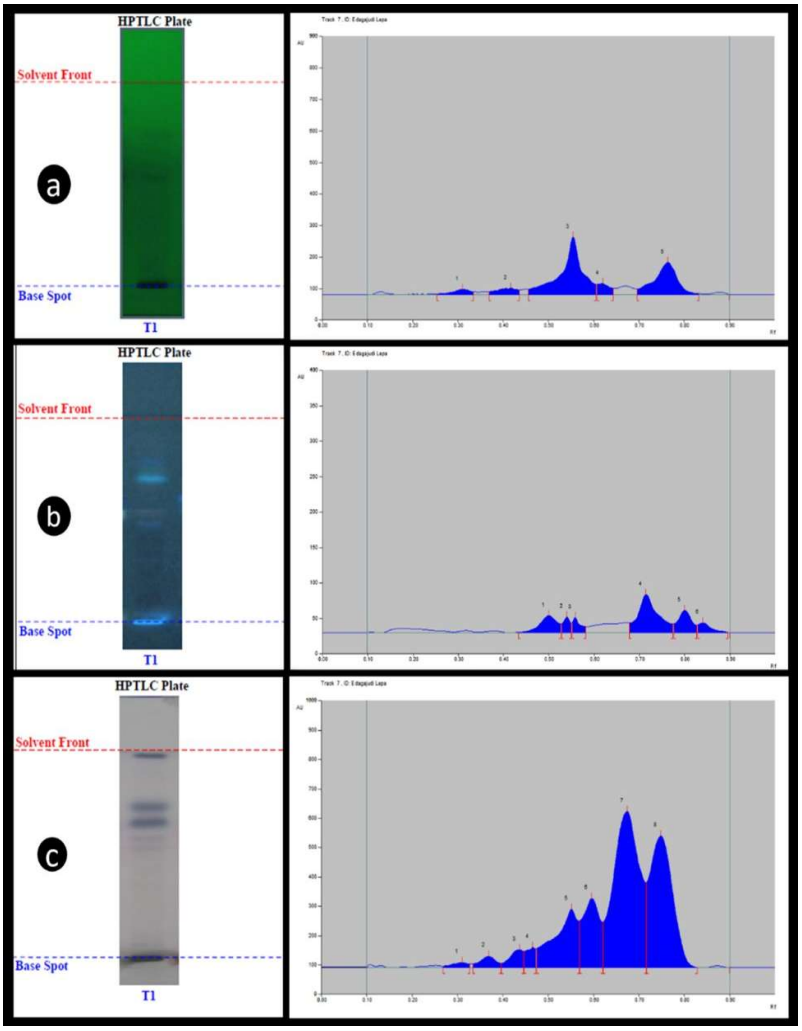

Figure 5: Photographs of HPTLC study of Edagajadi Lepa (a- HPTLC Chromatogram at UV 254nm, b- HPTLC Chromatogram at UV 366nm, c- HPTLC Chromatogram at UV 540nm)

Table 7: HPTLC Rf values of Edagajadi Lepa

Rf values at 254 nm		Rf values at 366 nm		Rf values at 540 nm	
0.31	Grey	0.50	Light Grey	0.31	Grey
0.42	Grey	0.55	Light Grey	0.37	Dark Blue
0.55	Dark Black	0.57	Grey	0.42	Purple
0.62	Dark Black	0.71	Grey	0.47	Grey
0.76	Grey	0.80	Sky Blue	0.55	Blue
		0.84	Light Green	0.62	Brown
				0.67	Dark Blue
				0.76	Light Blue

Complete analytical profile of Edagajadi Lepa

The complete analytical profile for one batch of Edagajadi Lepa is mentioned below in the **(Table 8)**.

Table 8: Analytical Profile of Edagajadi Lepa

Parameter	Observation	Inference
Organoleptic Evaluation	Sparsha(texture): Frictionless Roopa(colour): dark Brownish Gandha(smell): Odourless	Passes the test
Microscopic Characteristics	Lignified fibres of Edagaja, Oleoresin with crystals of Kushtha, Oil globules of Sarshapa, Resin content of Vidanga	Passes the test
Physico-chemical Evaluation		
pH Value	6	Slightly acidic
Loss on drying (%)	6.04%	-
Total Ash value (%)	18.68 %	-
Water soluble extract (% w/w)	59.30 %	-
Methanol soluble extract (%)	40.82 %	-
Petroleum ether (%w/w)	20.17 %	-
TLC value	TLC study shows 7 spots under 254 nm, 12 spots under 366 nm and 6 spots after spraying with Anisaldehyde sulphuric acid spray	-
HPTLC Value	HPTLC study shows Prominent peaks (Five at 254 nm, six at 366 nm and eight at 540 nm)	-

DISCUSSION

The present study, focused on the preparation and authentication of Edagajadi Lepa, a classical Ayurvedic formulation referenced in Charaka Samhita. The formulation comprises five herbal ingredients and one fermented preparation, all of which were properly identified, authenticated and processed in accordance with the Ayurvedic Pharmacopoeia of India **[15]**. The Sauveeraka (Fermented preparation) used in this formulation was prepared by using de-husked *Hordeum vulgare* Linn (Yava) with eight times of water and boiling till reduced half. Whole preparation was transferred to Earthen pot and lid was closed and kept undisturbed till fifteen days. After the completion of fermentation process, the lid was opened and filtration was carried out and used along with Edagajadi Lepa powder in the many skin conditions like Fungal infection (Dadru) and Psoriasis (Mandala Kustha) **[16]**.

Most ingredients in Edagajadi Lepa possess Tikta and Katu Rasa (bitter and pungent taste), Laghu, Ruksha and Tikshna Guna (light, dry and sharp qualities), Katu Vipaka (Pungent), Ushna Virya (hot potency), Kandughna (anti-pruritic), Kushthaghna (anti-dermatotic) and Krimighna (anti-microbial) properties **[17]**. which collectively contribute to its detoxifying and therapeutic actions. The constituents of this compound enter the Romakupa (hair follicles) and absorbed into Siramukha (terminal end of veins) and Swedavahi Srotasa (channels that transport sweat). This cutaneous biotransformation pacifies the Dosha (body humor) and breaks the Samprapti (pathogenesis) of Kustha Roga (skin diseases).

Pharmacognostic evaluation of the formulation revealed the presence of

Lignified fibres of *Cassia tora* (Edagaja), Oleoresin with crystals of *Saussurea lappa* (Kustha), Oil globules of *Brassica compestris* (Shweta Sarshapa), Resin content of *Embelia ribes* (Vidanga) confirming the authenticity and genuineness of the raw materials used. Physico-chemical analysis of Edagajadi Lepa further supported the pharmaceutical stability and purity of the compound. The pH value indicated its mild acidic nature, which is suitable for the external application on the skin. The reduced loss on drying (6.04%) suggests minimal content of moisture, which implies lower risk of microbiological contamination and better stability. The total Ash value (18.68 %) was found within acceptable limit, which indicates appropriate amount of inorganic content has been burned or oxidized. The water-soluble extractive value (59.30%) suggests a moderate presence of water-soluble contents such as organic acids, sugars and salts, while methanol soluble extract value (40.82 %) reflected the solubility of alcohol-based contents in the formulation. Petroleum ether value was found as (20.17 %), which suggests Edagajadi Lepa having significant amount of non-polar lipophilic phytoconstituents, which enhance absorption of drug through the skin and give better drug delivery at the site of local application **[18]**.

Chromatographic studies give further confirmation for the integrity of formulation. TLC (thin Layer Chromatography) analysis of Edagajadi Lepa suggest a common Rf value (0.18) across different wavelengths, indicates no separation of mixture or adulteration were found in the formulation. HPTLC fingerprint exhibited consistent RF values (0.55) across all wavelengths, confirming the presence of reproducible and stable bioactive compounds **[19]**. Overall, the Pharmacognostical and pharmaceutical findings from this study confirm that, Edagajadi Lepa is a scientifically validate and pharmaceutically stable compound. Analytical parameters obtained from this study serve as essential benchmarks for future reference.

Hence, and this analytical study contributes to the quality assurance of Ayurvedic formulations and supports their integration into modern medical practices through scientific validation and standardized procedures.

CONCLUSION

Edagajadi Lepa is polyherbal compound beneficial in many conditions such as Fungal infection (Dadru), Psoriasis (Mandala) and Helminthic (Krimi) diseases under the context of Kustha Roga (skin diseases). All the ingredients of this formulation were authenticated through Pharmacognostical evaluation. The physico-

chemical analysis provides a primary standard analytical profile of Edagajadi Lepa, which serves a reference for assess the quality of this formulation. Thus, present study describes essential analytical data that helps in confirming the identity, purity and standardization of the Edagajadi Lepa (polyherbal Ayurvedic formulation).

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Author Contributions: All authors have reviewed and approved the final version of the manuscript.

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