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Research Article

Pharmacological And Phytochemical Evaluation Of *Plectranthus Amboinicus* (Parnayavani) Using HPTLC And GC-MS: Insights into Traditional and Modern Applications

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ABSTRACT
Background: Plectranthus amboinicus, commonly known as Parnayavani, is traditionally used in Ayurvedic medicine to treat respiratory and digestive ailments. However, limited scientific validation exists to support its traditional claims.
Local Problem: Inadequate integration of traditional Ayurvedic uses with evidence-based modern pharmacological studies hinders its broader acceptance in healthcare.
Methods: This study employed High-Performance Thin Layer Chromatography (HPTLC) and Gas Chromatography-Mass Spectrometry (GC-MS) to analyze Parnayavani Arka prepared using classical Ayurvedic methods. Relevant phytochemical compounds were identified, and their pharmacological potential was evaluated.
Results: The analysis identified bioactive compounds, including thymol, linalool, and flavonoids, which align with the herb's traditional applications for respiratory and digestive conditions.
Conclusions: The findings validate the therapeutic potential of Parnayavani and highlight its potential for broader medicinal applications. Future research should focus on clinical trials and pharmacological validations.

Keywords: Plectranthus amboinicus, Parnayavani, HPTLC, GC-MS, Ayurveda, phytochemical analysis

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INTRODUCTION

Problem Description

Ayurveda extensively uses Plectranthus amboinicus, but its scientific validation remains underexplored, particularly in modern pharmacology.

Introduction

Plectranthus amboinicus (Lour.) Spreng., commonly referred to as Parnayavani in Sanskrit and known as Country Borage in English, is a member of the Lamiaceae family.,It is a perennial herb with a long history of use in Ayurvedic medicine.In Ayurveda this herb has many names like Chornika, Karpooravalli, Sugandhi, Parnika, Himasagar, and Ashmari bheda. Known for its Kaphavatahara properties, it has been utilized for treating respiratory and digestive issues, including cough, asthma, indigestion, and flatulence. The herb's properties—Tikta rasa (bitter taste), Kaṭu

vipaka (pungent post-digestive effect), and Uṣhna virya (heating potency)—align with its traditional usage in alleviating Kapha and Vata disorders. The herb's pharmacological properties, which include antiurolithiatic, antiepileptic, antiasthmatic, anti-inflammatory, antitumorigenic, analgesic, antioxidant, and antibacterial effects, are attributed to its rich content of phytoconstituents such as flavonoids, glycosides, phenols, tannins, and steroids. Among its active components, carvacrol and thymol, two volatile oils, are particularly noted for their beneficial effects on the respiratory system[[1]].

According to Ayurvedic principles, the leaves of *Plectranthus amboinicus* are acrid, bitter, aromatic, thermogenic, anthelmintic, lithontriptic, diuretic, stomachic, anodyne, appetizing, digestive, carminative, and potent for liver health. These properties make the herb effective in treating conditions such as nausea, vomiting, epilepsy, halitosis, bronchitis, renal and vesical calculi, hepatopathy, and malaria [[2]]. While the herb is not directly mentioned in Vedic or Samhita periods, it has been described by Acharya P. V. Sharma in the Priya Nighanṭu as *Coleus aromaticus* Benth., also known in South India as Kannikkoorkka or Panikkoorkka. This plant has long been used as a traditional remedy for fever, cough, asthma, and digestive issues [[3]].

Rationale

Bridging traditional and modern pharmacological knowledge could enhance its integration into contemporary healthcare systems. The identification of bioactive compounds supports these applications.

Specific Aims

This study aims to analyse Parnayavani Arka's phytochemical composition using HPTLC and GC-MS, providing evidence for its traditional uses.

MATERIALS AND METHODS

Objectives: To perform an extensive compilation and analysis in order to learn more about the therapeutic characteristics of *Parnayavani* (Plectranthus Amboinicus).

Context: The study was conducted at the Vasu Research Centre, Gujarat, utilizing standardized protocols for HPTLC and GC-MS analysis.

Intervention: Preparation: Parnayavani Arka was prepared via tri-distillation of Plectranthus amboinicus leaves following classical Ayurvedic methods.

Study of the Intervention: Approach: Phytochemical profiling was conducted using HPTLC and GC-MS to identify bioactive compounds.

Outcome Assessment: Identification of therapeutic compounds linked to traditional uses.

Pharmacological and medicinal uses of Parnayavani was compiled from classical and traditional books like Vaidya Tarakam, chikitsa manjiri etc further publications from different e-journals, along with publications such as Pharmacographia Indica, The Wealth of India, and Ayurvedic Pharmacopoeia of India.

Ethno-medicinal use

Table 1: Ethnomedicinal uses of Parnayavānī [4],[5],[6],[7],[8]

S. No.	Disease	Ethno - medicinal use	Method of Administration
1.	Diminished breast milk	In Indonesia, leaves are served as soup after delivery. North Sumatra's Batak tribes consume the leaf following childbirth.	Internal
2	As food	Chopped leaves for the stuffing's sage flavour. Eaten uncooked with butter and toast. used in the Philippines and Indonesia to cover up the smell of fish and goat meat Making "Bhajen" (cut leaves molded into pellets, dipped in a mixture made of chickpea flour, and then fried in butter)	Internal
3.	Urinary problems & Vaginal discharge	Leaves eaten raw	Internal
4.	Epilepsy, convulsions, asthma, bronchitis, sore throats, persistent coughs, and congestive heart failure	Infusion / decoction / syrups of leaf	Internal
5.	Centipede and Scorpion bite	Apply bruised leaf poultice	External
6.	Mouth corners that are cracked and have chapped lips	Application of leaf juice	External
7	Fever	A leaf lotion applied on the body	External
8.	Colic	It is advised to combine expressed juice with sugar or another appropriate vehicle and administer it.	Internal

9.	Conjunctivitis	Leaf juice applied in an expressive manner around the orbit	External
10.	Constipation	In Indonesia and Malaysia, juice from pounded leaf drank	Internal
11.	Influenza, cough, bronchitis and throat problems	Drink or bath with P. amboinicus juice / decoction	Internal/external
12.	Asthma	Juice or decoction prepared from leaves	Internal
13.	Cuts & Burns	On a flame, leaf paste is prepared and applied.	External
14.	Acute edematous otitis	Seed oil	External
15.	Diarrhea caused by a pathogen	Leaves taken with yogurt, buttermilk, or any other probiotic-containing foods	Internal

Uses of *Parụayavānī*

Table 2: *Parụayavānī* in *Keralīya granthās* [9],[10],[11]

S. No.	Disease	Therapeutic application / Formulation name	Kalpana	Method of Administration
1.	*Sarva vātaroga, Sannipāta jvara, Bhūtāveśa*	*Indrāni tailam*	*Taila*	Pana, Nasya,Lepa
2.	Fever with shivering	*Marica* pounded in *Parụayavānī*and *Vanatuḷasi* leaf juice	*Guḷika*	Internal
3.	In *Bāla*, *Jvaropadrava*	Porridge made with vacha, pippali, yashtimadhu, and candana paste, buttermilk, and *Parụayavānī* leaf juice	*Yavāgu*	Internal
4.	*Jvaropadrava* in *bāla*	Do *Talam* with the paste of *Kaṭurohini*, *Kuṣhṭaṃ*, and *Kriṣṇajīrakaṃ* is prepared by pounding it in *Parụayavānī* leaf juice and mixing it with breast milk.	*Kalka*	Talam
5.	*Vāyukṣobha* & Nīrvīzhcha	Oil Made from the juices of two pala Parụayavānī and Nirguụdi leaves, tila taila, and half as much eranda taila.	*Taila*	Siro abhyanga
6.	*Śvāsa Kāsa* in *bāla*	Parụayavānī, vāśa, bṛhati, kanṭakāri, puṣkaramūla, and other leaves are cooked, and the juice obtained is periodically served with karpūra and sugar candies.	*Svarasa*	Internal
7.	*Śvāsa* in children	*Putapāka svarasa* of *Parụayavānī, Tripādi, Vāśa, Tāmbūla* leaves with 1/4th breast milk with a small amount (Pinch) of *Lavanga, Jīraka*, and *Karpūra*	*Putapāka svarasa*	Internal
8.	*Śvāsa*&*Kāsa* in children	Decoction of *Parụayavānī* leaves	Kvātha	Internal
9.	*Śvāsa*&*Kāsa* in children	Oil with *Parụayavānī*leaves	Taila	Siro abhyanga
10.	*Jvara*	*Jīraka* Leaf Decoction	*Kvātha*	Internal
11.	*Jvara, Kāsa, Śvāsa*	Decoction of leaves with *Jīraka* and *Pippali*	*Kvātha*	Internal
12.	*Villain chuma* or *Nilamkari chuma*	*Kāsasaṃhāri leha* *Kāsamarda avaleha*	*Leha*	Internal

TAXONOMICAL DERIVATION

Kingdom : Plantae
Order : Lamiales
Family : Lamiaceae
Genus : Coleus
Species : Coleus Amboinicus

Initial Steps and Evolution

The preparation of Parnayavani Arka followed traditional Ayurvedic methods, yielding a clear, aromatic liquid. Analytical methods were optimized for accurate profiling.

Process Measures

HPTLC identified key phytochemical constituents with Rf values matching known standards.
GC-MS provided detailed compound profiles, revealing bioactives such as thymol, linalool, and flavonoids.

PHYTOCHEMICAL ANAALYSIS

Sample of Parnayavani It hails from Kerala, India, the species' most well-known habitat. The sample was submitted to phytochemical analysis after drug authentication, and the results were as follows:

Table 3. Physico-Chemical Analysis Report of Sample P. Amboinicus

SL NO.	PARAMETERS	VALUE
1	DESCRIPTION COLOUR ODOUR TASTE CONSISTEBLITY	TRANPSRENT CLEAR LIQUID AROAMTTIC BITTER LIQUID
2	pH(5% Aqueous)	6.9
3	Refractive Index at 40°	1
4	Specific gravity	0.85
5	Aflatoxins	Absent
6	Test for Heavy metals	Absent
7	Flavonoids	++
8	Fixed volatile content	++
9	TLC Report	Rf values: 0.10,0.22,0.53

HPTLC Analysis

HPTLC was performed on silica gel 60 F_{254} plates using toluene:ethyl acetate:acetic acid (7:3:0.1 v/v) as mobile phase. Chromatograms were recorded at wavelengths of 254 nm, 366 nm and 540 nm. The resulting fingerprints showed significant peaks corresponding to different phytoconstituents.

GC-MS Analysis

GC-MS was performed using a Perkin Elmer Clarus 500 system with an Elite 5ms capillary column. The column was operated under the following conditions:
Injection temperature: 290°C
Ion source temperature: 150°C
Carrier gas: Helium (1 mL/min)
Oven temperature: Initially 50°C, ramped to 280°C at 8°C/min.
The mass spectra were analysed and showed the presence of various volatile and non-volatile compounds.

Phytochemical analysis

Following conventional protocols, phytochemical experiments using HPTLC were conducted at the Vasu Research Centre, Makarpura, Vadodara-390010, Gujarat, India.

Table: 4- HPTLC FINGERPRINTING REPORT (METHODOLOGY)	
Test solution Preparation: In an evaporating dish, 50 g of the sample was precisely weighed and evaporated until it was completely dry. 1 mL of methanol was added to it. Next, filtered with 0.22 micron assistance. The resulting filtrate was then utilized for HPTLC fingerprinting.	
Making the spray reagent (sulphuric acid reagent – anisaldehyde): 10 mL of glacial acetic acid is combined with 0.5 mL of Anisaldehyde, 85 mL of methanol, and 5 mL of sulphuric acid (98%) in order of concentration	
Chromatographic Conditions:	
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	15 µL
Distance Between Tracks	0.0 mm
Development Mode	CAMAG TLC Twin Trough Chamber
Chamber Saturation Time	30 minutes
Mobile Phase (MP)	Toluene : Ethyl acetate : Acetic acid (7 : 3 : 0.1 v/v)
Visualization	@ 254 nm, @ 366 nm and @ 540 nm (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± 50C for 3 minutes

Table No 5. Phytochemical constituents of Parnayavani Arka

Sr. No.	**Parameters**	**Rf Value**
1	Linalool	0.33
2	Terpene	0.40
3	Eugenol	0.47

4	Monoterpene	0.62
5	Flavonoids	0.53
6	β - ansarone	0.77
7	Kaempferol	0.29
8	Flavanoid Glycoside	0.57
9	Carvacrol	0.70
10	Thymol	0.88

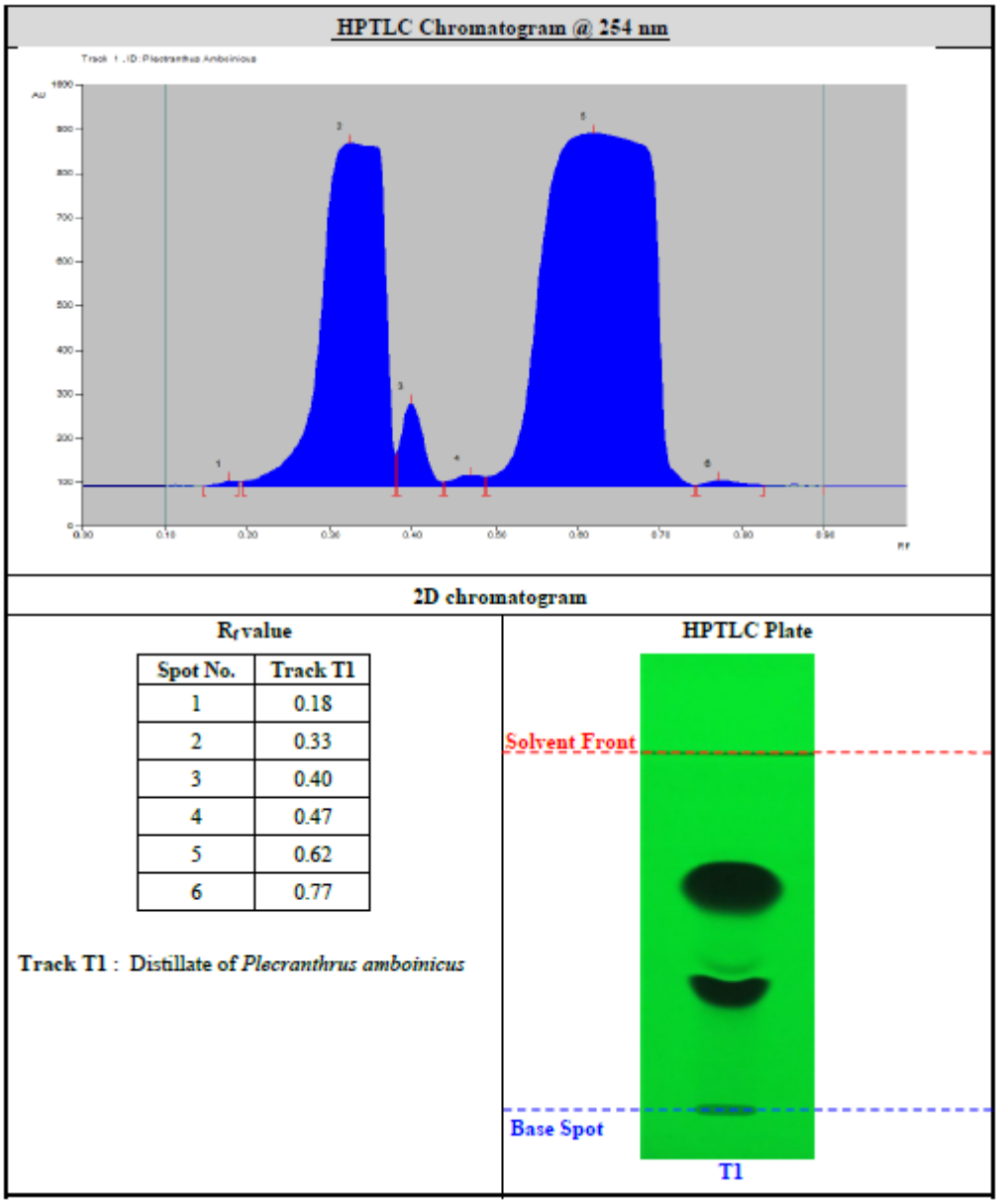

Figure.1 Banding pattern and Rf values at 254 nm on an HPTLC plate

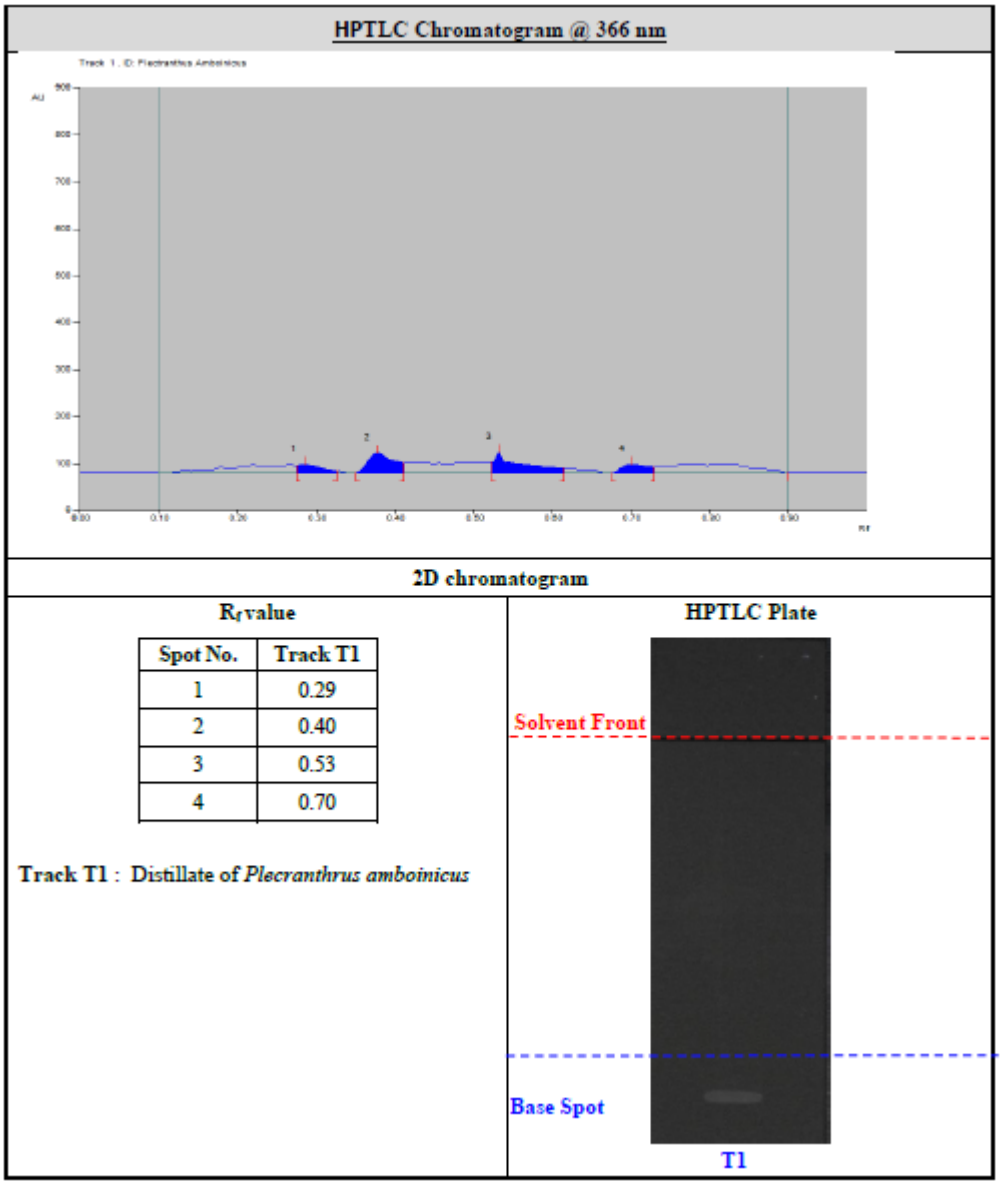

Figure 2: Banding pattern and Rf values at 366 nm on an HPTLC plate

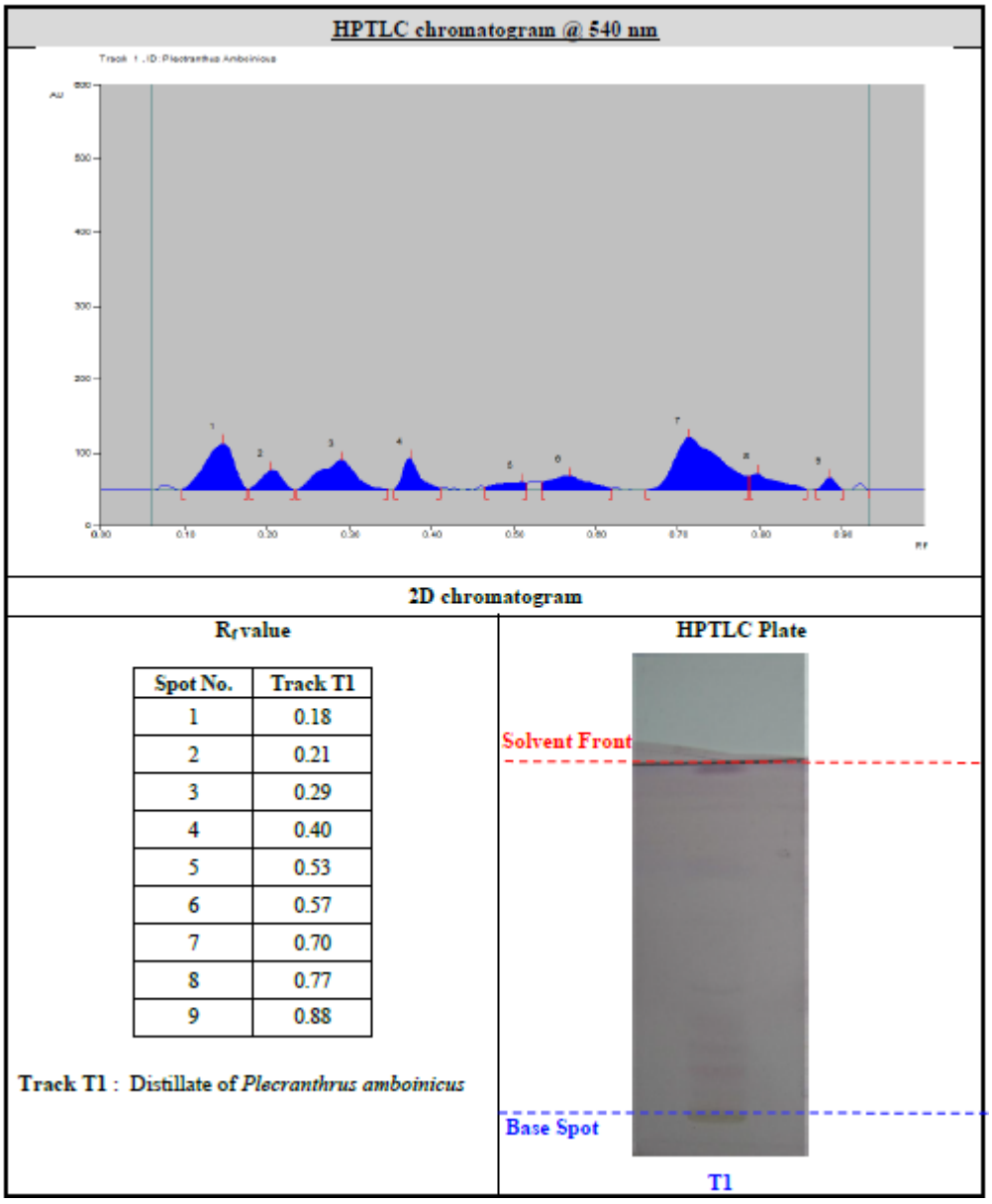

Figure 3: Banding pattern and Rf values at 540 nm on an HPTLC plate

RESULTS AND DISCUSSION

Organoleptic and Physico-chemical Analysis

The organoleptic properties and physico-chemical properties of "Parnayavani Arka" are displayed in Table 1. The distillate, called Arka, is described as a clear, translucent liquid with an aromatic scent and a bitter flavor. The acquired drug had a refractive index of 1 and a specific gravity of 0.85. The pH of the drug is 6.9.

Phytochemical Tests

The Parnayavani Arka contained linalool, terpene, eugenol, monoterpene, flavonoids, B-onsarone, kaempferol, flavonoid glycoside, glycoside, and thymol, according to the results of the phytochemical screening (Table 3). Numerous biological actions have been described for the majority of the known phytochemical substances, including

Linalool

A flowery and pungent terpene alcohol is called linalool. It is found in more than 200 plants, including citrus fruits and lavender [12]. anticonvulsant, antidepressant, muscle relaxant, anti-anxiety, and stress reliever [13].The infiltration of inflammatory cells and mucus hypersecretion in the lung tissues were inhibited by linalool [14]. Some research suggests that linalool may have bronchodilator effects, meaning it may help to widen the airways in the lungs. This could potentially be useful in conditions where airway constriction is a problem, as chronic obstructive pulmonary disease (COPD) or asthma.

Terpene

Terpenes or isoprenoids are the most abundant and diverse class of naturally occurring substances found mainly in plants[15]. They are responsible for giving plants their taste, aroma and color. Terpenoids have been shown to have anti-allergic, antiparasitic, antiviral, anti-

inflammatory, antibacterial, blood sugar lowering, antispasmodic, antifungal and immunostimulant properties. These properties may be useful in the prevention and treatment of various diseases, such as cancer [16]. Studies in humans have also shown that volatile terpene molecules have positive effects on asthma. α-Pinene has been shown to be bronchodilator in volunteers, which means it could be useful for asthma patients.[17]

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Monoterpene

The tiniest terpene kind is called a monoterpene. Essential oils, fragrances, and a number of structural isomers 21 are known to have them as their main component. They have the composition C10H16 and are derived from different plants, fruits, and flowers. The efficacy of bronchodilation in Wistar rats and guinea pigs has been shown in several animal experiments [18]. Antiviral, antibacterial (anti-tuberculosis), anticonvulsant, anti-inflammatory, anti-Alzheimer, antidepressant, and anti-Parkinsonian actions are only a few of the characteristics of monoterpenes. According to one study, exposure to monoterpenes and asthmatic teenagers may have a statistically significant localized airway impact that is unique to monoterpenes [19].

Eugenol

As a phenolic monoterpenoid, eugenol (EUG) is a versatile naturally occurring chemical that is commonly found in essential oils from a broad variety of plant species [20]. Numerous illnesses, including those pertaining to the reproductive system, the neurological system, cholesterol and blood sugar, microbial infections, cancer, inflammations, hypertension, and digestive problems, can be effectively treated with EUG [21]. Eugenol's effectiveness as a bronchodilator was demonstrated in the research "Antiasthmatic effect of eugenol (4-allyl-2-methoxyophenol) through bronchodilatory and immunomodulatory propertiess" [22].

Flavonoids

Flavonoids, a class of phytochemicals found in many fruits, vegetables, and leaves, may be useful in medicinal chemistry. Flavonoids are known for their antiviral, anticancer, and antioxidant properties, among other health benefits. Furthermore, they possess cardio- and neuroprotective qualities. The kind of flavonoid, its mode of action (if any), and its bioavailability all impact these physiological processes.[23] Flavonoids have the potential to be beneficial and helpful in delaying the onset of lung disorders such asthma, ARDS, lung cancer, and COPD, according to Lago et al. (2014).[24]

β -ansarone

Numerous pharmacological properties, including anti-inflammatory, anticancer, antiapoptotic, antioxidant and neuroprotective activities, are demonstrated by β-asarone.[25] Asarone has broncholytic, expectorant and antimicrobial properties.[26]

Kaempferol

Kaempferol is structurally comparable to the hormone estrogen, as it belongs to the flavonoid family. Kaempferol is found in fruit and vegetables such as apples, grapes, tomatoes, broccoli, pine trees and ginkgo leaves. Kaempferol has anti-inflammatory, antibacterial, antioxidant, cardiovascular and neuroprotective properties. Kaempferol is similar to the hormone estrogen and can be used to treat malignancies that are affected by hormones, including leukemia, ovarian, breast, cervical and hepatocellular carcinomas.[27]

Carvacrol

For Carvacrol, Known for antimicrobial, antifungal, and antioxidant properties. Carvacrol has a notable medicinal role in treating respiratory ailments..[28]

Thymol

Thyme's rich flavor, lovely aroma, and powerful antibacterial effects are attributed to a white, crystalline component called thymol (2-isopropyl-5-methylphenol).[29] The main active ingredient in thyme essential oil is thymol, which has been shown to have antiseptic, antibacterial, antifungal, anthelmintic, antiviral, antioxidant, expectorant, antispasmodic, carminative, diaphoretic, sedative, antirheumatic and even anticancer, antihyperlipidemic and antihyperglycemic effects.[30],[31],[32],[33],[34],[35]

High Performance Thin Layer Chromatography

Following derivatization, the HPTLC photo documentation of "Parnayavani Arka" (Figs. -1, 2, 3) revealed six, four, and nine spots under 254 nm, 366 nm, and 540 nm, respectively. Spot Rf value 0.40 was often found using each of the three detection techniques. Commonly found spots with Rf values of 0.18,0.29,0.53, and 0.70 were found using any two detection techniques. Since all three techniques produced the best possible band separation, they may all be utilized as HPTLC fingerprint patterns to determine the mixture's composition (Figs. 1, 2, 3). Two high peaks and one peak, corresponding to four distinct chemicals in the methanol extract with Rf values of 0.33, 0.62, and 0.40, were found by densitometry scanning at 254 nm (Fig. 1). There were two high and two low peaks at 366 nm, which corresponded to four distinct chemicals in the methanol extract; the main peak found had an Rf value of 0.40, 0.53, 0.29, and 0.70 (Fig. 2). Two High Peaks and seven peaks with an Rf value of 0.18 and 0.70, respectively, were found at 540 nm.

Overview of GC-MS Analysis

The GC-MS analysis of Plectranthus amboinicus (Purnayavani Arka) was conducted to identify the key chemical compounds present. The major retention times (RT) identified were at 15.894 min, 18.665 min, 21.626 min, and 24.052 min, each representing a distinct set of chemical components. Below is a detailed breakdown of the identified compounds and their relevance, especially in respiratory conditions.

Identified Compounds and Their Properties [36],[37],[38]

RT: 15.894 min

1. Thymol (C10H14O) - CAS 89-83-8
2. Phenol, 2-methyl-5-(1-methylethyl)- (C10H14O) - CAS 499-75-2
3. p-Cymen-7-ol (C10H14O) - CAS 536-60-7
4. Phenol, p-tert-butyl- (C10H14O) - CAS 98-54-4

Key Active Compounds:

- Thymol: Known for its antiseptic, antifungal, and antibacterial properties, thymol is widely used in traditional medicine for treating infections and respiratory conditions.
- p-Cymen-7-ol: It has antioxidant and antimicrobial properties, enhancing the medicinal benefits of the plant.

RT: 18.665 min

1. 1,2-Ethanediamine, N-(2-aminoethyl)- (C4H13N3) - CAS 111-40-0
2. 1H-Cyclopenta[c]furan-3(3ah)-one (C10H12O4) - CAS 900141-74-4
3. Meglumine (C7H17O5N) - CAS 6284-40-8
4. 1,3,5-Trioxepane (C4H8O3) - CAS 5981-06-6

Key Active Compounds:

- 1,2-Ethanediamine, N-(2-aminoethyl)-: Used as a stabilizer or chelating agent in medicinal formulations.
- Meglumine: Known for its use in radiographic contrast media, not a direct bioactive agent but part of the stabilizing agents.

RT: 21.626 min

1. Silanes, Tetramethyl- (C4H12Si) - CAS 75-76-3
2. 1,3-Dioxolane-2-methanol (C4H8O3) - CAS 5694-68-8
3. Penicillamine, 3TMS derivative (C14H35O2NSSi3) - CAS 167029-28-9

Key Active Compounds:

- Silanes, Tetramethyl-: Primarily used as protective agents in organic compounds.
- 1,3-Dioxolane-2-methanol: Used in chemical reactions, not directly bioactive.

RT: 24.052 min

1. 1,2-Ethanediamine, N-(2-aminoethyl)- (C4H13N3) - CAS 111-40-0
2. Malonic acid, bis(2-trimethylsilylethyl ester) (C13H28O4Si2) - CAS 90744-45-9
3. Tartronic acid, 3TMS derivative (C12H28O5Si3) - CAS 38165-93-4

Key Active Compounds:

- Malonic acid: Used in synthetic applications, intermediate rather than bioactive.

Active Chemical Compounds in Respiratory Conditions

Based on the analysis, several key compounds are beneficial for respiratory conditions:

1. Thymol (C10H14O): Known for its antiseptic, antifungal, and antibacterial properties, particularly useful in respiratory treatments for bronchitis, coughs, and congestion.
2. p-Cymen-7-ol (C10H14O): An antioxidant and antimicrobial agent that aids in protecting the respiratory system from infections.
3. Phenol, 2-methyl-5-(1-methylethyl)- (C10H14O): Exhibits anti-inflammatory and antimicrobial effects, helpful in managing respiratory infections and inflammation.

Discussion

The phytochemical analysis of Parnayavani Arka supports its traditional use in Ayurveda for treating respiratory and digestive ailments. Compounds like linalool and thymol, identified in the GC-MS analysis, are known for their bronchodilatory effects, which align with the herb's traditional application in treating asthma and cough. The presence of flavonoids and eugenol further validates the herb's use for its anti-inflammatory and antimicrobial effects.

Conclusion

Linalool, terpene, eugenol, monoterpene, flavonoids, B-onsarone, kaempferol, flavonoid carvacrol, and thymol were found in the extract of Parnayavani Arka, according to preliminary phytochemical tests. These compounds are reportedly bioactive and may contribute to the nebulization's therapeutic effect. The Parnayavani Arka formulation's HPTLC fingerprint profile may be utilized for quality assurance and verification. In the GC-MS analysis of Plectranthus amboinicus (Purnayavani Arka) confirms the presence of important bioactive compounds such as Thymol and p-Cymen-7-ol. These compounds are beneficial for respiratory conditions, primarily due to their antimicrobial, anti-inflammatory, and antioxidant properties. Thus, it can be said that Parnayavani Arka may be evaluated using these factors. Future research on Parnayavani Arka may use this paper as a reference.

Implications

Further studies, including clinical trials, are needed to establish its efficacy and safety.

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