

Pharmacognostical Standardization and Phytochemical Characterization of *Phyllanthus Emblica*, *Hypericum Perforatum*, And *Calophyllum Inophyllum* for Development of a Polyherbal Formulation

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Abstract:

Pharmacognostical standardization and preliminary phytochemical characterization of *Phyllanthus emblica* (dried fruits), *Hypericum perforatum* (aerial parts), and *Calophyllum inophyllum* (leaves) were carried out to evaluate their quality, authenticity, and therapeutic potential for development of a polyherbal formulation. Organoleptic studies confirmed characteristic color, odor, taste, and texture, while physicochemical analyses, including total ash, water-soluble ash, acid-insoluble ash, moisture content, and foreign matter, demonstrated good quality and proper processing. Preliminary phytochemical screening revealed that *Phyllanthus emblica* is rich in glycosides, tannins, phenolics, flavonoids, saponins, and carbohydrates; *Hypericum perforatum* contains alkaloids, flavonoids, and phenolics; and *Calophyllum inophyllum* possesses saponins, flavonoids, tannins, and steroids/triterpenoids. These complementary phytochemical profiles support their traditional uses and justify their combination in a polyherbal formulation. The study provides essential evidence for standardization, safety, and efficacy, forming a foundation for future formulation development and pharmacological evaluation.

Keywords: *Phyllanthus emblica*, *Hypericum perforatum*, *Calophyllum inophyllum*, pharmacognosy, phytochemical screening, polyherbal formulation

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1. Introduction

Medicinal plants have been integral to traditional healthcare systems due to their diverse bioactive compounds and therapeutic properties (Chandel, Pathak & Tailang, 2011). Their incorporation into scientifically validated polyherbal formulations requires rigorous pharmacognostical and phytochemical standardization to ensure quality, safety, and efficacy (Gaire & Subedi, 2014).

***Phyllanthus emblica* L.** (amla or Indian gooseberry) is widely recognized in Ayurveda for its rich phytochemical profile, including vitamin C, phenolics, flavonoids, and tannins, contributing to antioxidant, anti-inflammatory, antimicrobial, anti-diabetic, hepatoprotective, and anticancer activities (Krishnaveni & Mirunalini, 2010; Begum et al., 2024; Prananda et al., 2023). Experimental studies have demonstrated its ability to modulate oxidative stress, enhance immune responses, and promote tissue regeneration, supporting its traditional use as a Rasayana (Gaire & Subedi, 2014; Ma et al., 2024; Srirama et al., 2012).

***Hypericum perforatum* L.** (St John's Wort) contains bioactive constituents such as hypericin,

hyperforin, and flavonoids that exhibit antidepressant, antiviral, antibacterial, anti-inflammatory, and neuroprotective effects (Barnes, Anderson & Phillipson, 2001; Nathan, 1999; Miller, 1998; Russo et al., 2014). Clinical and preclinical studies confirm its efficacy in mild to moderate depression and highlight potential therapeutic roles in neurodegenerative, dermatological, and viral infections (Suryawanshi et al., 2024; Galeotti, 2017; Tausova et al., 2022; Liu et al., 2014).

***Calophyllum inophyllum* L.** (tamanu) has been traditionally used for wound healing, skin disorders, inflammation, and infections. Scientific investigations have demonstrated its anti-inflammatory, antioxidant, anticancer, and tissue regenerative properties, largely due to flavonoids, saponins, and phenolic compounds (Ruangsuriya et al., 2023; Shanmugapriya et al., 2017; Yimdjo et al., 2004). Studies further indicate its ability to modulate reactive oxygen species, inhibit inflammatory mediators, and promote cellular repair mechanisms, validating its ethnomedicinal use (Islam et al., 2019; Lima & Nunes, 2020).

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Considering the complementary phytochemical profiles and therapeutic potential of these three plants, their combination in a polyherbal formulation may provide synergistic pharmacological benefits. Comprehensive pharmacognostical, physicochemical, and phytochemical evaluations are essential to establish reliable quality parameters, ensure safety, and support the rational development of the formulation for clinical applications (Begum et al., 2024; Prananda et al., 2023; Suryawanshi et al., 2024).

2. Collection of Plant Material

The plant materials, including fruits of *Phyllanthus emblica*, aerial parts of *Hypericum perforatum*, and seeds of *Calophyllum inophyllum*, were collected from a nearby botanical garden during their optimal season. The collected materials were washed with distilled water, shade-dried at room temperature to preserve phytoconstituents, and then powdered using a mechanical grinder. The powdered samples were stored in airtight containers in a cool, dry place until further extraction.

3. Organoleptic Properties

Organoleptic properties are the sensory characteristics of a plant material evaluated through sight, smell, taste, and touch. They help in the preliminary identification and quality assessment of the sample.

Color, odor, taste (bitter, sweet, sour, or astringent), and texture (powdery, coarse, oily, etc.) are observed to detect authenticity, purity, and possible contamination. This simple evaluation serves as a quick, non-instrumental quality control step before further chemical and pharmacological analysis.

4. Soxhlet extraction

Soxhlet extraction is a conventional technique used to extract bioactive compounds from solid materials through continuous solvent reflux and condensation. In saffron, ethanol-based Soxhlet extraction effectively isolates key constituents such as crocins, safranal, and other active compounds.

Materials:

The materials required for Soxhlet extraction included high-purity ethanol as the extraction solvent, a Soxhlet extractor assembly consisting of a round-bottom flask and condenser, and a heating mantle to maintain continuous reflux. Filter paper and glass wool were used to hold and secure the powdered sample within the extraction chamber. A distillation setup and rotary evaporator were employed for efficient solvent recovery and concentration of the extract. An analytical balance was used for accurate weighing of the plant material prior to extraction.

Procedure:

A weighed amount of plant material was placed in a Soxhlet extractor thimble connected to a round-bottom flask and condenser, and ethanol was used as the solvent for efficient extraction of phytoconstituents. The system was heated,

allowing the solvent to vaporize, condense, and continuously percolate through the material over several hours. After extraction, the solution was concentrated with a rotary evaporator, filtered to remove residues, and stored in a dark, airtight container, ensuring preservation of light-sensitive compounds and adherence to safety precautions.

5. Macroscopic studies

The selected crude drugs were subjected to studies organoleptic characters viz., color, odour, appearance, taste, texture etc.

5.1 Physicochemical Evaluation

5.1.1 Determination of Ash Value

Ash content is an important physicochemical parameter that represents the inorganic residue remaining after complete combustion of organic matter. It serves as an indicator of the total mineral content, particularly inorganic salts, present in a sample and is widely used to assess its quality and purity.

Materials and Equipment:

The determination of ash content required a heat-resistant crucible for incineration, a muffle furnace (550–600°C) for complete combustion, a desiccator to cool the sample without moisture absorption, and an analytical balance for precise weighing of the sample and ash residue.

Procedure:

A representative sample was accurately weighed and transferred into a preheated crucible, which was cooled in a desiccator before use. The crucible with the sample was gradually heated in a muffle furnace at 550–600°C until complete combustion, yielding constant-weight ash. After cooling in a desiccator to prevent moisture absorption, the crucible was weighed to determine the ash content.

Calculation:

$$\text{Ash Content (\%)} = \frac{\text{Weigh of Ash}}{\text{Weight of Sample}} \times 100$$

5.1.2 Determination of Ash Value

This comprehensive procedure ensures the precise determination of total ash content, considering both naturally occurring minerals and added impurities.

Materials and Equipment:

The determination required a heat-resistant crucible or ashing dish made of porcelain, quartz, or metal to withstand high temperatures. A muffle furnace (550–600°C) was used for incineration, while a desiccator helped cool the sample without moisture absorption. An analytical balance was employed for accurate weighing of the sample and the resulting ash.

Procedure:

A representative sample was accurately weighed and transferred into a preheated crucible, evenly spread to ensure uniform combustion. The crucible was heated gradually in a muffle furnace at 550–600°C until complete ashing. After cooling in a desiccator to prevent moisture absorption, the

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crucible was weighed, and the residue represented the total ash content.

Calculation:

$$\text{Total Ash Content (\%)} = \frac{\text{Weight of Ash}}{\text{Weight of Sample}} \times 100$$

5.1.3 Determination of acid insoluble ash

Materials and Equipment:

The materials used included a heat-resistant crucible or ashing dish (porcelain, quartz, or metal) for sample incineration, a muffle furnace (550–600°C) for complete combustion, a desiccator for cooling without moisture absorption, and an analytical balance for accurate weighing. Dilute hydrochloric acid (HCl) was also used to dissolve acid-soluble salts for determining acid-insoluble ash.

Procedure:

A measured sample was first subjected to total ash determination until complete incineration. The resulting ash was treated with dilute hydrochloric acid to dissolve acid-soluble matter and gently heated. The mixture was filtered through ashless paper to collect the insoluble residue, which was washed with hot water, dried, and incinerated in a muffle furnace at 550–600°C until a constant weight was reached. After cooling in a desiccator, the remaining residue represented the acid-insoluble ash.

Calculation:

$$\text{Acid Insoluble Ash Content (\%)} = \frac{\text{Weight of Acid Insoluble Ash Content}}{\text{Weight of Sample}} \times 100$$

5.1.4 Determination of water soluble ash

Materials and Equipment:

The materials required for the determination of water-soluble ash included a heat-resistant crucible or ashing dish made of porcelain, quartz, or metal for incineration. A muffle furnace (550–600°C) was used for ashing the sample. A desiccator was employed to cool the crucible without moisture absorption, and an analytical balance ensured accurate weighing. Distilled water was used to dissolve the water-soluble components of the total ash during the procedure.

Steps:

A weighed sample was first subjected to total ash determination until complete incineration. The resulting ash was treated with distilled water to dissolve water-soluble components and filtered through ashless paper to collect the insoluble residue. The residue was washed, dried, and incinerated in a muffle furnace at 550–600°C until constant weight. After cooling in a desiccator, the remaining residue was weighed, and the difference from the total ash represented the water-soluble ash.

Calculation:

$$\text{Water-Soluble Ash Content (\%)} = \frac{\text{Water-Soluble Ash Content}}{\text{Weight of Sample}} \times 100$$

5.1.5 Determination of loss on drying

Loss on drying (LOD) is a method used to determine the moisture content present in a sample. It is widely applied in pharmaceutical, food, and material analysis to evaluate product stability, quality, and suitability for storage and processing.

Materials and Equipment:

The materials required for loss on drying determination included an analytical balance for accurate weighing of the sample, a drying oven or moisture analyzer to remove moisture by controlled heating, and a desiccator to cool the dried sample and protect it from atmospheric moisture before final weighing.

Steps:

A representative sample was accurately weighed and dried either in a pre-weighed dish at 105–110°C using an oven or according to the moisture analyzer's programmed conditions. After drying, the sample was cooled in a desiccator to prevent moisture absorption and reweighed. The weight loss after drying represented the loss on drying.

Calculation:

$$\text{Loss on Drying (\%)} = \frac{\text{Initial Weight} - \text{Final Weight}}{\text{Initial Weight}} \times 100$$

5.1.6 Moisture content

Moisture content is a crucial parameter in various industries, including food, pharmaceuticals, and materials testing. Accurate determination of moisture content is essential for quality control, product development, and process optimization. Here's a general guide on the procedure for determining moisture content.

Materials and Equipment:

The determination of moisture content required an analytical balance for accurate weighing of the sample, a drying oven or moisture analyzer to remove moisture under controlled heating conditions, and a desiccator to cool the dried sample and prevent reabsorption of atmospheric moisture before final weighing.

Procedure:

A representative amount of the sample was accurately weighed using an analytical balance. The sample was then dried either in a hot air oven at 105–110°C until a constant weight was obtained or by using a moisture analyzer as per instrument guidelines. After drying, the sample was cooled in a desiccator to prevent moisture absorption and reweighed. The decrease in weight indicated the moisture content.

6. Preliminary phytochemical analysis of extracts

Preliminary phytochemical analysis is the initial step used to identify the major chemical

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constituents present in plant extracts. It helps detect bioactive compounds that contribute to the therapeutic properties of medicinal plants and provides a foundation for further detailed chemical and pharmacological studies.

6.1 Test for Alkaloids:

Alkaloids were identified using standard qualitative tests. A solution of the plant extract was prepared and treated separately with different reagents. The formation of characteristic precipitates indicated the presence of alkaloids.

- **Mayer's test:** Formation of a yellow or cream precipitate.
- **Dragendorff's test:** Appearance of an orange or reddish-brown precipitate.
- **Wagner's test:** Development of a reddish-brown precipitate.
- **Hager's test:** Formation of a yellow or orange precipitate.

Positive results in these tests confirmed the presence of alkaloids in the extract.

6.2 Test for Carbohydrates:

Carbohydrates were detected using standard qualitative tests. The plant extract solution was treated with specific reagents, and characteristic color changes or precipitate formation indicated their presence.

- **Molisch's test:** Formation of a violet ring at the junction after adding Molisch's reagent and concentrated sulfuric acid.
- **Fehling's test:** Appearance of a brick-red precipitate upon heating with Fehling's solution.
- **Benedict's test:** Development of a colored precipitate ranging from green to brick-red after heating with Benedict's reagent

Positive reactions confirmed the presence of carbohydrates in the extract.

6.3 Test for Glycosides:

Glycosides were identified using the modified Borntrager's test. The plant extract was mixed with chloroform and then treated with dilute ammonia. The appearance of a pink, red, or violet color in the ammoniacal layer indicated the presence of glycosides in the extract.

6.4 Test for Phytosterols and Triterpenoids:

Phytosterols and triterpenoids were detected using standard qualitative tests. The extract was dissolved in chloroform and treated with specific reagents to observe characteristic color changes.

- **Liebermann test:** Color change from violet to blue or green indicated a positive result.
- **Liebermann-Burchard test:** Formation of a green color confirmed the presence of sterols or triterpenoids.

- **Salkowski test:** Development of a red-brown color suggested the presence of these compounds.

6.5 Test for Protein and Amino Acids:

Proteins and amino acids were identified using standard qualitative tests.

- **Millon's test:** Addition of Millon's reagent followed by heating produced a brick-red precipitate, indicating the presence of proteins.
- **Ninhydrin test:** Treatment of the extract with ninhydrin resulted in a blue or purple color, confirming amino acids.
- **Biuret test:** Addition of Biuret reagent produced a violet color, indicating the presence of proteins.

Positive color changes confirmed the presence of proteins and amino acids in the extract.

6.6 Test for Phenolic and Tannins:

Phenolic compounds and tannins were detected using standard qualitative tests.

- **Ferric chloride test:** Addition of a few drops of ferric chloride to the extract produced a blue-black or green coloration, indicating the presence of phenolics or tannins.
- **Lead acetate test:** Formation of a white or yellow precipitate after adding lead acetate solution confirmed the presence of tannins.

Positive color changes or precipitate formation indicated the presence of phenolic compounds in the extract.

6.7 Test for Flavonoids:

Flavonoids were identified by the following qualitative test:

- **Shinoda test:** Addition of Shinoda reagent to the extract resulted in the development of a red or pink color, indicating the presence of flavonoids.

The appearance of the characteristic red coloration confirmed flavonoids in the extract.

6.8 Test for Oils and Fats:

Oils and fats were detected using the following qualitative test:

- **Oily spot test:** A small amount of the extract was applied to filter paper. The formation of a translucent, permanent greasy spot indicated the presence of oils and fats.

The appearance of the translucent spot confirmed the presence of lipids in the extract.

6.9 Test for Saponins:

Saponins were detected using the foam test.

- **Foam test:** The extract was shaken vigorously with water. The formation of persistent froth or stable foam indicated the presence of saponins.

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Stable foam formation confirmed the presence of saponins in the extract.

Materials Needed:

Ethanollic extracts of *Phyllanthus emblica*, *Hypericum perforatum*, and *Calophyllum inophyllum* were used for suspension preparation. A 0.5% carboxymethyl cellulose (CMC) solution served as the suspending vehicle, with 0.1% Tween 80 added to improve dispersion. Distilled water was used for dilution. The formulation required a beaker, magnetic stirrer or glass rod, measuring syringes or graduated cylinders, and amber glass bottles for storage.

Procedure:

The suspension was prepared by dissolving 0.5 g of CMC in 100 mL of distilled water with continuous stirring until a homogeneous 0.5% solution was obtained. Tween 80 (0.1 mL per 100 mL) was optionally added to improve dispersion. The required dose of extract (200 mg/kg) was calculated based on the body weight and number of rats, and the accurately weighed extract was gradually incorporated into the CMC-Tween solution with continuous stirring to obtain a uniform, lump-free suspension. The final preparation was stored in amber-colored bottles at 4°C and shaken well before administration.

7. Result & Discussions

7.1 Macroscopic studies:

Organoleptic evaluation is an important preliminary step in the identification and quality assessment of herbal raw materials. It provides rapid information about plant characteristics using sensory parameters such as color, odor, taste, and texture. The following observations were recorded for the selected plant parts:

1. *Phyllanthus emblica* (dried fruits/powder):

The dried fruit pieces were irregular and wrinkled, showing a brown to dark brown color after powdering. The odor was slight and characteristic with a faint acidic note, while the taste was distinctly sour and astringent. No significant foreign organic matter was observed, indicating good quality and cleanliness of the material.

2. *Hypericum perforatum* (aerial parts/powder):

The powdered aerial parts were fine, containing fragments of leaves and flowers with a green to yellowish-green color. The odor was aromatic and slightly balsamic, and the taste was bitter with mild pungency. No significant foreign matter was observed, indicating good quality and proper processing.

3. *Calophyllum inophyllum* (leaves):

The leaves were broad, ovate to oblong, measuring 6–15 cm long and 3–6 cm wide, with a green to dark green color. They had a slightly aromatic to musty

odor and a bitter, astringent taste, indicating flavonoid and tannin content.

No foreign matter was detected, confirming the sample's purity.

Conclusion:

The organoleptic characteristics of all three plant samples matched pharmacopeial and ethnobotanical descriptions, providing a key step in the preliminary authentication and standardization of raw materials prior to physicochemical and phytochemical analyses.

Table 1: Organoleptic characters of plants:

S. No	Parameters	Observations of <i>Phyllanthus emblica</i> dried fruits/powder	Observations of <i>Hypericum perforatum</i> aerial parts/powder	Observations of <i>Calophyllum inophyllum</i> leaves
1.	Shape	Irregular, wrinkled pieces / powder	Fine powder or fragmented leaves and flowers	Broad, ovate or oblong leaves
2.	Size	Powder / ≤ 1 cm fruit segments	Powder / leaf fragments ≤ 1 cm	Leaves ~6–15 cm long, ~3–6 cm wide
3.	Odour	Slight, characteristic, faintly acidic	Aromatic, slightly balsamic	Slightly aromatic to musty
4.	Taste	Astringent, sour	Bitter, slightly pungent	Bitter, astringent
5.	Colour	Brown to dark brown powder	Green to yellowish-green powder	Green to dark green (fresh or dried)
6.	Foreign organic matter	Absent / within permissible limits ($<2\%$)	Absent / within permissible limits	Absent / within

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7.2 Physicochemical Standardization of Proposed Plants

Standardization is essential for ensuring the quality, purity, safety, and efficacy of herbal materials. Pharmacognostical and physicochemical analyses help confirm plant identity and detect adulteration. In this study, three medicinal plants—Phyllanthus emblica (dried fruit), Hypericum perforatum (aerial parts), and Calophyllum inophyllum (leaves)—were evaluated for key parameters, including ash values, foreign matter, water-soluble ash, acid-insoluble ash, and moisture content.

• Total Ash Value

Total ash indicates the overall inorganic content in plant material, including both natural minerals and external contaminants. In this study, Hypericum perforatum showed the highest ash value (7.8%), likely due to its mineral-rich aerial parts, followed by Calophyllum inophyllum (6.3%), and Phyllanthus emblica (5.2%), reflecting lower inorganic content. All values fall within acceptable limits, supporting the authenticity and quality of the samples.

• Foreign Organic Matter

Foreign organic matter refers to unwanted materials such as other plant parts, soil, insects, or fungal elements. WHO and Indian Pharmacopoeia guidelines set the permissible limit at <2%. In this study, all three plants complied: Hypericum perforatum (0.8%), Phyllanthus emblica (0.9%), and Calophyllum inophyllum (1.1%). These low values indicate careful collection and processing, ensuring high quality and minimal contamination.

• Water-Soluble Ash

Water-soluble ash measures the portion of total ash that dissolves in water, mainly representing water-soluble salts and electrolytes. Hypericum perforatum had the highest value (3.2%), indicating a richer content of soluble minerals, possibly associated with its flavonoids and phenolics. Phyllanthus emblica and Calophyllum inophyllum showed 2.3% and 2.1%, respectively. All values are within safe limits, confirming minimal soluble impurities.

• Acid-Insoluble Ash

Acid-insoluble ash indicates siliceous matter, such as sand, which may arise from poor washing or drying on dusty surfaces. Phyllanthus emblica showed the lowest value (0.4%), reflecting thorough cleaning, while Hypericum perforatum and Calophyllum inophyllum had slightly higher values (0.6% and 0.7%). All results are within pharmacopoeial limits, confirming hygienic processing suitable for medicinal use.

• Moisture Content

Moisture content influences the stability and shelf life of herbal materials, with excess water

promoting microbial growth and degradation. In this study, Phyllanthus emblica, Hypericum perforatum, and Calophyllum inophyllum had moisture levels of 8.5%, 9.5%, and 10.2%, respectively, all below the 12% limit. The slightly higher moisture in Calophyllum inophyllum leaves may reflect their thicker structure. Overall, these plants show suitable organoleptic and physicochemical properties, supporting their use in herbal formulations and traditional applications.

Table 2: Standardization parameters

S. No	Parameters	Phyllanthus emblica dried fruits/powder	Hypericum perforatum aerial parts/powder	Calophyllum inophyllum leaves
1	Ash value	5.2%	7.8%	6.3%
2	Foreign organic matter	0.9%	0.8%	1.1%
3	Water soluble ash	2.3%	3.2%	2.1%
4	Acid insoluble ash	0.4%	0.6%	0.7%
5	Moisture content	8.5%	9.5%	10.2%

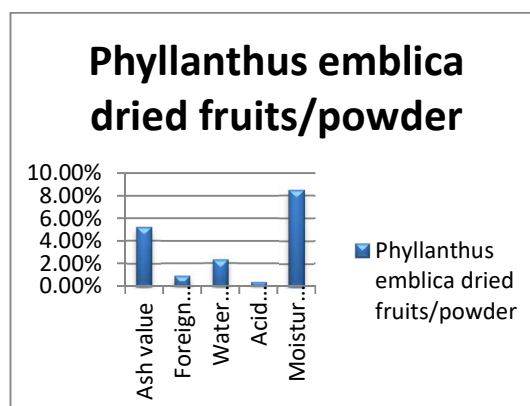


Figure 1: Phyllanthus emblica dried fruits/powder

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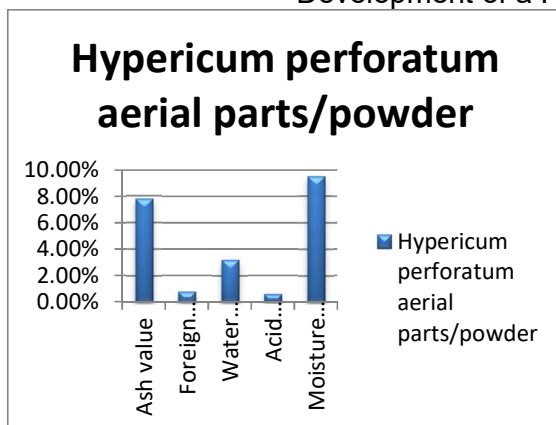


Figure 2:Hypericum perforatum aerial parts/powder

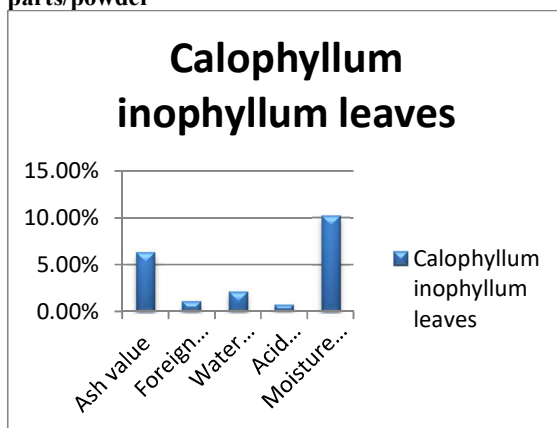


Figure 3:Calophyllum inophyllum leaves

7.3Preliminary Phytochemical Analysis of Extracts:

Preliminary phytochemical screening is essential to identify key bioactive compounds in medicinal plants. Detecting classes like alkaloids, flavonoids, glycosides, steroids, tannins, saponins, and carbohydrates helps predict biological activity. In this study, Phyllanthus emblica (fruits), Hypericum perforatum (aerial parts), and Calophyllum inophyllum (leaves) were tested for these secondary metabolites.

1. Steroids and Triterpenoids

All three plants tested positive in the Liebermann–Burchard and Salkowski tests, confirming the presence of steroids and triterpenoids. These bioactive compounds, known for anti-inflammatory, hepatoprotective, and antimicrobial effects, may contribute to the therapeutic potential of these plants, especially in wound healing and skin-related treatments.

2. Saponins

Saponins were present in Phyllanthus emblica and Calophyllum inophyllum, as shown by the foam test, but absent in Hypericum perforatum. These compounds are linked to immune stimulation, cholesterol regulation, and enhanced bioavailability of other constituents, while their absence in

Hypericum perforatum reflects a different phytochemical profile.

3. Alkaloids

Alkaloids, identified by Hager's and Mayer's tests, were found only in Hypericum perforatum, supporting its documented mood-regulating and antidepressant effects. Their absence in Phyllanthus emblica and Calophyllum inophyllum indicates that these plants' pharmacological activities rely on other phytochemical classes.

4. Glycosides

Glycosides were detected only in Phyllanthus emblica through Borntrager's and Keller–Killiani tests, confirming the presence of anthraquinone and cardiac glycosides. These compounds may contribute to its antioxidant and hepatoprotective effects. Their absence in the other two plants suggests a different phytochemical profile with limited involvement in cardiovascular or laxative activities.

5. Tannins and Phenolics

All three plants showed the presence of phenolic compounds via Gelatin and Ferric Chloride tests. This is consistent with Phyllanthus emblica's well-known richness in tannins and vitamin C. These phenolics confer antioxidant, astringent, and antimicrobial properties, supporting their use in wound healing and anti-aging applications.

6. Flavonoids

Flavonoids were detected in all three plants using Ferric Chloride and Alkaline Reagent tests. These polyphenols provide antioxidant, anti-inflammatory, and vascular-protective effects, supporting the traditional use of these plants against oxidative stress and inflammation.

7. Proteins

Proteins were detected in all three plants using Biuret and Xanthoproteic tests. While not directly pharmacologically active, these proteins can aid tissue regeneration, making them beneficial in wound healing and topical applications.

8. Carbohydrates

Phyllanthus emblica tested positive for Fehling's test, confirming the presence of reducing sugars, which may contribute to its nutritional and rejuvenating (Rasayana) properties. The other two plants showed no detectable carbohydrates, likely due to lower sugar content or extraction differences.

Conclusion

Phyllanthus emblica showed a wide range of phytochemicals, Hypericum perforatum was rich in flavonoids, alkaloids, and phenolics, and Calophyllum inophyllum contained saponins, flavonoids, and tannins. These findings support their traditional medicinal uses and potential pharmacological applications.

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Table 3: Phytochemical Profile of Phyllanthus emblica, Hypericum perforatum, and Calophyllum inophyllum leaves:

S. No	Chemical Tests	<i>Phyllanthus emblica</i> dried fruits/powder	<i>Hypericum perforatum</i> aerial parts/powder	<i>Calophyllum inophyllum</i> leaves
1	Tests for Steroids and Triterpenoids:			
	• Lieberman-Burchard Test	+	+	+
	• Salkowski Test	+	+	+
2	Test for Saponins:	+	–	+
	• Foam Test	+	–	+
3	Tests for Alkaloids:			
	• Hager's Test	–	+	–
	• Mayer's Test	–	+	–
4	Tests for Glycosides:			
	• Borntraeger's Test	+	–	–
	• Keller-Killiani Test	+	–	–
5	Tests for Tannins and Phenolics:			
	• Gelatin Test	+	+	+
	• Ferric Chloride Test	+	+	+
6	Tests for Flavonoids:			
	• Ferric Chloride Test	+	+	+

	• Alkaline Reagent Test	+	+	+
7	Tests for Proteins:			
	• Biuret Test	+	+	+
	• Xanthoproteic Test	+	+	+
8	Test for Carbohydrates:			
	• Fehling's Test	+	–	–

8. Conclusion

The present study successfully standardized and characterized *Phyllanthus emblica* (dried fruits), *Hypericum perforatum* (aerial parts), and *Calophyllum inophyllum* (leaves) through organoleptic, physicochemical, and preliminary phytochemical evaluations. Organoleptic assessments confirmed characteristic sensory features, while physicochemical analyses, including ash values, moisture content, and foreign matter, demonstrated good quality and proper processing of all three plants. Phytochemical screening revealed a diverse profile: *Phyllanthus emblica* was rich in glycosides, tannins, phenolics, flavonoids, saponins, and carbohydrates; *Hypericum perforatum* contained alkaloids, flavonoids, and phenolics; and *Calophyllum inophyllum* showed saponins, flavonoids, tannins, and steroids/triterpenoids. These findings align with their traditional medicinal uses and suggest complementary therapeutic potential, justifying their combination in a polyherbal formulation. Overall, this study establishes essential pharmacognostical and phytochemical evidence for the quality, safety, and efficacy of these plants, providing a strong foundation for the development of standardized polyherbal products for medicinal or nutraceutical applications, with scope for further quantitative and pharmacological investigations.

9. Acknowledgements

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10. Conflict of Interest

The authors declare that they have no conflict of interest.

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