

# Pharmacognostical and Phytochemical Analysis of *Ipomoea mauritiana* Jacq. Tuber

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## ABSTRACT

**Introduction:** The purpose of this investigation was to undertake a thorough pharmacognostical and phytochemical analysis of the tuber of *Ipomoea mauritiana* Jacq. to determine the diagnostic features and to study the occurrence of bioactive constituents. **Methodology:** Tubers were harvested, identified, washed, dried, and powdered. Macroscopic and microscopic examinations were carried out to record physical and anatomical features. Physicochemical parameters including moisture content, ash value, swelling index, and extractive values with different solvents were measured. Ethanol extracts performed phytochemical screening to determine whether secondary metabolites were present. **Results:** Macroscopic observation showed brown-colored tubers with a wrinkled, rough surface and fibrous texture. Calcium oxalate crystals, starch grains, and cork cells were detected by microscopic and powder analysis. According to physicochemical examination, the ethanol extractive value was 8.5% w/w, the moisture content was 3.4% w/w, the total ash was 4.5% w/w, and the acid-insoluble ash was 1.1% w/w. The presence of saponins was indicated by the foaming index, which was determined to be >100. While proteins and tannins were found to be absent, phytochemical screening revealed the presence of carbohydrates, flavonoids, phenols, terpenoids, alkaloids, and glycosides. **Conclusion:** The investigation is able to successfully set the pharmacognostic standards and emphasize the therapeutic value of *Ipomoea mauritiana* tubers as a result of the presence of pharmacologically active compounds. This proves its usage as per traditional medicine and creates a necessary baseline data for standardization, quality control, and other pharmacological studies.

**Keywords:** *Ipomoea mauritiana*, Macroscopy, Microscopy, Phytochemical screening, Tuber.

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## INTRODUCTION

Herbal remedies are most popular, being used by approximately 80% of the world's population. Their popularity can be understood due to their attributed safety, efficiency, cultural compatibility, and fewer side effects compared to traditional pharmaceuticals; also, they are considered economical and accessible. Herbal medicinal products may be beneficial therapeutically, yet numerous others are also causative of adverse effects and interactions with allopathic drugs, like traditional pharmaceuticals. There is an especially high potential for interaction between the use of herbal remedies and traditional drugs for drugs that have therapeutic windows that are narrow.<sup>1</sup>

*Ipomoea mauritiana* Jacq. (Figure 1) is a pharmaceutical species under the family Convolvulaceae, and it is known to be employed in both traditional folk medicine and Ayurveda. One

of the main sources of Vidari, an Ayurvedic medication that is an ingredient in roughly 50 Ayurvedic formulations, including Chyawanprash, is this plant.<sup>2</sup> It is a glabrous, twining shrub that has a very large tuberous root and palmate, lobed leaves up to 8 cm in length. *Ipomoea mauritiana* is sometimes found in semi-evergreen forests, mainly in open areas. This plant's tubers are very valuable in traditional medicine due to their healing aspects.<sup>3</sup> Several intriguing chemicals, such as taraxerol, taraxerol acetate, umbelliferone,  $\beta$ -sitosterol, scopoletin, and 7-O- $\beta$ -d-glycopyranosyl scopoletin (also called Scopolin), were extracted from the tubers using methanol. These roots are used as a stimulant, carminative, tonic, galactagogue, and to increase hunger. They also support rejuvenation therapy. It's interesting to note that the alcohol extract from the tubers has a restorative effect on the central nervous system by having both stimulating and depressive effects.<sup>4</sup> The quality of botanical specimens and scientific study depend greatly on the comprehensive morphological, anatomical, and microscopic descriptions. In addition to identifying plants, these characteristics are utilised to find contaminations, adulterations, and substitutes in plant products.<sup>5</sup> Many nations have used microscopy to identify herbal products, as documented in various pharmacopoeias, due to its benefits, which include low sample size requirements, rapid



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output, dependability, ease of use, and affordability.<sup>6</sup> Adulteration or substitution is all about swapping out the original plant material for a different variety or intentionally adding something foreign to boost the weight or potency of a product, or even to cut costs. A common problem arises from misidentification, which often leads to the wrong use of herbal medicine or natural products. One frequent issue is that a single common name might be used for two or more completely different species. Thankfully, these challenges can be tackled through pharmacognostic research on medicinal plants.<sup>7</sup> The qualities of the chemical components in the crude medicine that are responsible for certain physiological effects on the human body are disclosed by the extractive values.<sup>8</sup>

## METHODOLOGY

### Collection, authentication and drying of tuber

The plants used in this study were carefully collected from Rayirath Gardens, a well-known wholesale plant nursery located in the Pananchery Panchayath of Thrissur. They were also verified by Dr. Ranjusha, a botanist from the Department of Botany at NSS College in Ottapalam.

The tubers were well cleaned under running tap water to flush out adhering soil particles. The tubers were then cleaned with distilled water to get rid of any remaining impurities. The cleaned tubers were then cut into uniform pieces to ensure uniform drying. The slices were then dried under shade in a well-ventilated area with protection from direct sunlight to help maintain their natural color and volatile compounds.

### Macroscopic evaluation

Organoleptic evaluation of *Ipomoea mauritiana* tuber was done in terms of colour, odour, size, shape, texture, fracture, and surface features.

### Microscopic evaluation

#### TS of *Ipomoea mauritiana* tuber

Thin section of tuber is taken stained with Safranin, mounted in glycerin.

#### Preparation of powder

The cleaned tubers were then cut into uniform pieces to ensure uniform drying. The slices were then dried under shade in a well-ventilated area with protection from direct sunlight to help maintain their natural color and volatile compounds and powdered with help of grinder.

#### Powder microscopy

Powder is stained with safranin. Mounted in glycerin and observed through the microscope. Repeated the procedure in 2-3 slides to get maximum characters.

## Physiochemical evaluation

### Moisture content

The Loss on drying method, which is standardised in accordance with pharmacopoeias, was used to assess the moisture content of *Ipomoea mauritiana* powder.

A flat weighing plate that had been previously weighed and dried was filled with 2-5 g of precisely weighed *Ipomoea mauritiana* powder. For 3-4 hr, the test sample was dried at  $105^{\circ}\text{C} \pm 2^{\circ}\text{C}$  in a hot air oven. The sample was dried and then allowed to cool in a desiccator for about half an hour before being weighed. Until a consistent weight was achieved (the difference between two consecutive weighing was less than 0.5 mg), drying was done in 30-min increments, with cooling and weighing in between. The provided formula was used to calculate the moisture content.

$$\text{Moisture Content (\%)} = \left( \frac{\text{Initial weight} - \text{Final weight after drying}}{\text{Initial weight}} \right) \times 100$$

Care was taken to maintain a temperature such that overheating was avoided, thereby preventing active components from being degraded. Triplicate measurements were made to assure accuracy.<sup>9</sup>

### Swelling index

A 100 mL measuring cylinder was filled with precisely weighed *Ipomoea mauritiana* powder, weighing around 1.0 g. The cylinder was filled with 25 mL of distilled water. For 1 hr, the mixture was shaking violently every 10 min. After shaking, the cylinder was left undisturbed for 3-4 hr at room temperature ( $25 \pm 2^{\circ}\text{C}$ ). It was noted how much volume the swelled substance ultimately occupied. The formula was used to calculate the Swelling Index.<sup>10,12</sup>

The Swelling Index is determined as the volume in mL of space taken up by the swollen material per gram of test sample.<sup>11</sup>

$$\text{Swelling Index} = \frac{\text{Volume occupied by swollen material}}{\text{Weight of sample (g)}}$$

### Foreign Matter Determination

About 100 g of *Ipomoea mauritiana* powder was weighed carefully and spread thinly on a clean, flat surface (preferably white paper or tray). The sample was visually inspected to sort out any foreign organic matter (e.g., other plant parts, weeds, etc.), foreign inorganic matter (like stones, soil, sand), or any extraneous material not part of *Ipomoea mauritiana*. The foreign material was removed manually and weighed on a precision balance. The percentage of foreign matters was calculated according to the formula.<sup>10,12</sup>

$$\text{FOM(\%)} = \left( \frac{\text{Weight of Foreign Organic Matter}}{\text{Total Sample Weight}} \right) \times 100$$

## Total Ash

A silicon crucible that had been previously burned and tarred was filled with roughly 2-3 g of precisely weighed *Ipomoea mauritiana* powder. To demonstrate the lack of carbon, the material was burned in a muffle furnace by gradually increasing the temperature to 450–600°C until it turned white. The substance was incinerated to prevent flaming. After cooling in a desiccator, the crucible was weighed. In relation to the air-dried powder, the percentage of total ash was calculated.<sup>10,12</sup>

$$\text{Total Ash (\%)} = \left( \frac{\text{Weight of ash}}{\text{Weight of air-dried sample}} \right) \times 100$$

## Determination of Acid-Insoluble Ash

In a muffle furnace, 2–3 g of precisely weighed *Ipomoea mauritiana* powder was fired in a silica crucible that had been heated and tarred to 450–600°C until it turned white and was determined to be carbon-free. To determine the entire amount of ash, the crucible was dried up and weighed. After adding 25 mL of diluted hydrochloric acid (70 g/L) to the total amount of ash, it was gently heated for 5 min. After filtering the insoluble material through ashless filter paper and washing it with hot water until the filtrate was neutral, it was once more ignited in a muffle furnace set to 450–600°C until a consistent weight was achieved. The crucible was weighed after cooling in a desiccator. The air-dried powder was used to calculate the percentage of acid-insoluble ash.<sup>10,12</sup>

$$\text{Acid-Insoluble Ash (\%)} = \left( \frac{\text{Weight of acid-insoluble ash}}{\text{Weight of air-dried sample}} \right) \times 100$$

## Water soluble ash

In a muffle furnace, 2-3 g of carefully weighed *Ipomoea mauritiana* powder was fired in a silica crucible that had been heated and tarred to 450-600°C until it turned white, indicating the lack of carbon. To determine the entire amount of ash, the crucible was cooled in a desiccator and weighed. After adding 25 mL of distilled water to the total amount of ash, the mixture was gently heated for 5 min. After washing with hot water and filtering the insoluble portion using ashless filter paper, the filtrate was moved to a conical flask. After being dried and burned at 450-600°C in a muffle furnace, the insoluble portion that remained on the filter paper was cooled in a desiccator and weighed. The weight of the entire amount of ash was subtracted from the weight of the residue. The amount of water-soluble ash was the weight difference. The percentage of water-soluble ash was calculated in comparison to the air-dried powder.<sup>10,11</sup>

$$\text{Water-Soluble Ash (\%)} = \left( \frac{\text{Weight of total ash} - \text{Weight of insoluble ash}}{\text{Weight of air-dried sample}} \right) \times 100$$

## Determination of extractive value

In a glass stoppered conical flask, combine 5 g of coarsely powdered air-dried material with 100 mL of solvent.

After 6 hr of frequent shaking, it was left to stand for 18 hr.

Quickly filtered. The filtrate (25 mL) was evaporated. The percentage of air-dried material that contained extractable materials was determined.

Solvent used: n Hexane, Petroleum ether, chloroform, acetone, ethyl acetate, ethanol.

$$\text{Extractive Value (\%)} = \left( \frac{\text{Weight of extract after evaporation}}{\text{Weight of air-dried sample}} \right) \times 100$$

## Phytochemical studies

### Extraction

A Soxhlet equipment was used to pack 25 g of coarsely powdered tuber. Extracted with 500 mL of 80% ethanol. Next, Whatman No. 1 filter paper was used to filter the extract. Evaporation-concentrated and kept in an airtight container.

## Phytochemical screening of plant extract

### Test for Alkaloids

#### Mayer's Test

Started by adding 2 mL of Mayer's reagent to 2 mL of the extract. Almost immediately, a creamy precipitate appeared, which indicated that alkaloids were present.<sup>13-15</sup>

#### Hager's Test

Introduced 1 to 2 mL of Hager's reagent to 2 mL of the extract. This time, a yellow precipitate formed, hinting at the presence of alkaloids.

#### Wagner's Test

Added 1 to 2 mL of Wagner's reagent to 2 mL of the extract. A reddish-brown precipitate emerged, confirming the presence of alkaloids.



Figure 1: *Ipomoea mauritiana*.

## Test for Carbohydrates

### Molisch's Test

To start, mix 2 mL of Molisch's reagent with 1 mL of your test solution and give it a good shake. Next, carefully add one ml of concentrated Sulphuric Acid (H<sub>2</sub>SO<sub>4</sub>) along the side of the test tube. If you see a violet ring where the two layers meet, congratulations! That's a sign that carbohydrates are present.

### Fehling's Test

For this test, boil 1 mL of your test solution together with 1 mL each of Fehling's solution A and B in a water bath. If a red precipitate forms at the bottom, it indicates that reducing sugars are present.

### Benedict's test

In this test, combine 2 mL of Benedict's reagent with 2 mL of your test solution and boil it in a water bath. If a red precipitate appears, it confirms the presence of reducing sugars.

## Test for Flavonoids

### Ferric Chloride Test

Added a few drops of ferric chloride solution to 2 mL of the test solution. Almost instantly, a vibrant green color appeared, signaling the presence of flavonoids.

### Shinoda's Test

In 2 mL of the extract, we mixed in 10 drops of dilute Hydrochloric Acid (HCl) along with a small piece of magnesium turning. This reaction produced a pink, reddish, or brown hue, hinting at the presence of flavonoids.

Aqueous Sodium Hydroxide Test: We introduced a few drops of 10% aqueous sodium hydroxide (NaOH) to 2 mL of the extract.

The formation of a yellow precipitate confirmed the presence of flavonoids.

## Test for Glycosides

### Baljet's Test

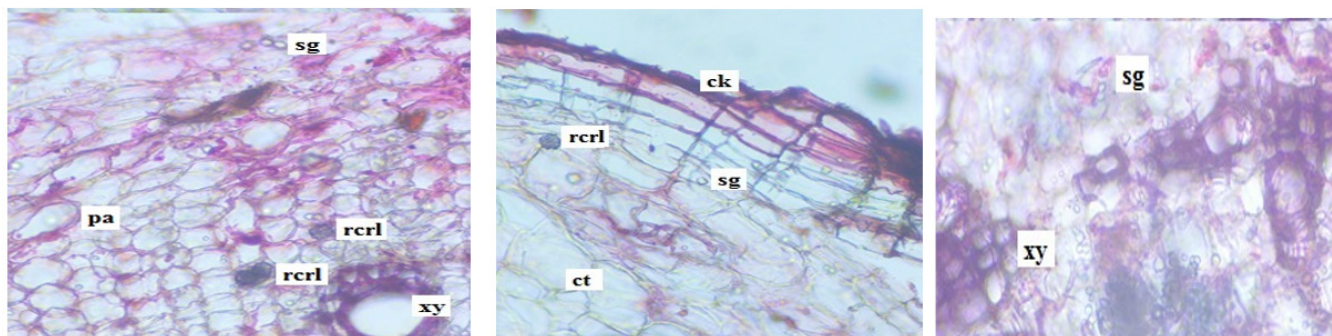
Added 2 mL of sodium picrate solution to about 2-3 mL of the sample. We observed a color change from yellow to orange, which confirmed that glycosides were present.

### Bortrager's Test

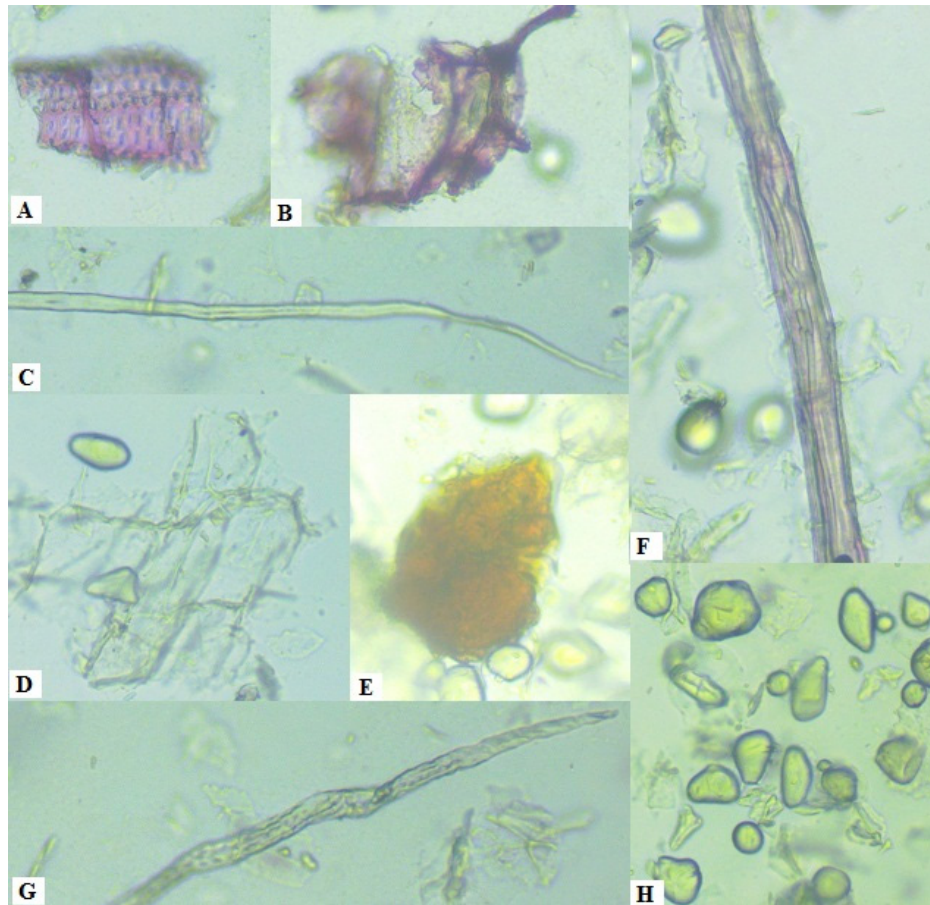
In this test, we mixed 2 mL of the sample solution with water and chloroform. Once the layers separated, we collected the organic (chloroform) layer and shook it with some dilute ammonia. The lower chloroform layer turned a deep red, while the upper layer shifted from green to blue and violet, indicating the presence of anthraquinone glycosides.

**Table 1: Physicochemical constants of tuber of *Ipomoea mauritiana*.**

Sl. No.	Physico-chemical constants	Result
1	Moisture content	3.4% w/w
2	Ash values	
	a) Total ash	4.5%w/w
	b) Acid insoluble ash	1.1%w/w
	c) Water soluble ash	3.8%w/w
3	Foreign organic matter	Less than one
4	Swelling index	3 mL
5	Foaming index	>100
6	Extractive values	
	n Hexane	1.0
	Petroleum Ether	1.3
	Chloroform	4.0
	Acetone	7.5
	Ethyl Acetate	2.0
	Ethanol	8.5



**Figure 2:** *Ipomoea mauritiana* tuber thin section: ck.: cork; ct.: cortex; pa.: parenchyma; rcrl.: calcium oxalate rosette crystals; sg.: starch grains; xy.: xylem.



**Figure 3:** Powder microscopy: A.: bordered pitted vessel; B.: cork cells; C.: non-lignified fiber; D.: parenchyma cells; E.: colored content; F.: lignified fiber; G.: tracheid; H.: starch grains.

## Test for Phenolics and Tannins

### Ferric Chloride Test

Added a few drops of a 5% ferric chloride solution to the aqueous extract. Almost immediately, a dark green or bluish-black color emerged, which confirmed the presence of phenolics or tannins.

### Lead Acetate Test

Next, mixed 1 mL of lead acetate solution with 2 mL of the test solution. A thick white precipitate formed, indicating that tannins were present.

## Test for Proteins and Amino Acids

### Millon's Test

2 mL of the extract and mixed it with 2 mL of Millon's reagent, then boiled the mixture. A white precipitate formed, which changed to red when heated, indicating that proteins were present.

### Ninhydrin Test

Added two drops of Ninhydrin solution to 2 mL of the extract and heated it in a water bath. A violet color emerged, suggesting that amino acids were present.

**Table 2: Phytochemical screening.**

Sl. No.	Constituents	Presence of constituents in Ethanol
1	Carbohydrate	++
2	Alkaloid	+
3	Flavonoids	++
4	Proteins	-
5	Glycoside	+
6	Phenols	++
7	Tannins	+
8	Terpenoids	++

## Test for Saponins

### Foam or Froth Test

To start, two ml of the extract and mixed it with 20 mL of distilled water in a graduated cylinder. After giving it a good shake for about 15 min, we looked for froth formation, which indicated that saponins were present.

## Test for Terpenoids

### Salkowski's Test

For this test, we dissolved two ml of the extract in chloroform and then added an equal amount of concentrated sulphuric acid. We observed that the chloroform layer turned red, while the acid layer displayed a greenish-yellow fluorescence, confirming the presence of terpenoids.

## Quantitative Estimation of Phytoconstituents

### Total phenolic content: Folin-Ciocalteu method

Began by preparing a gallic acid solution in methanol at different concentrations (20, 40, 60, 80, and 100 µg/mL). For each concentration, add 1 mL of gallic acid to a 10 mL standard flask. Then, add 5 mL of 10% Folin-Ciocalteu reagent and 4 mL of 7% sodium carbonate to get a total volume of 10 mL. Shake the blue mixture thoroughly and then placed it in a water bath at 40°C for 30 min. The absorbance at 760 nm was then measured against a blank, ensuring that every experiment was conducted in triplicate. Our calibration curve was then plotted using the average absorbance readings from the various gallic acid concentrations. A concentration of 100 µg/mL of extract was generated for the sample. 1 mL of the extract was taken, and 5 mL of 10% Folin-Ciocalteu reagent and 4 mL of sodium carbonate were added.<sup>16</sup>

### Total Flavonoid Content: Aluminium Chloride Colorimetric Method

Standard quercetin at concentrations of 20, 40, 60, 80, and 100 µg/mL was prepared. A 10 mL volumetric flask containing 4 mL of double-distilled water was filled with 1 mL of quercetin in methanol for each concentration. 0.3 mL of 5% sodium nitrite was introduced first, followed by 0.3 mL of 10% aluminium chloride after 5 min, then 2 mL of 1M sodium hydroxide at the 6

min mark. 2.4 mL of double-distilled water was then added to get the total amount down to 10 mL, and everything was thoroughly mixed. A blank containing all the chemicals except quercetin was used to test the absorbance of the resultant pink mixture at 510 nm. Our calibration curve was made using the average absorbance readings from the various quercetin concentrations. Prepared the plant extract using the same protocol as usual.<sup>17</sup>

## RESULTS AND DISCUSSION

### Macroscopic evaluation

The tuber is dark brown to brown outside and creamy white inside, with a typical slightly aromatic smell, an elongated or irregularly cylindrical tuberous shape (30–34 cm long and 9–10 cm wide), a rough, uneven surface with wrinkles, ridges, and rootlet scars, a smooth mucilaginous fracture when fresh, and a moist, fibrous texture when fresh which becomes short and mealy on drying.

### Microscopic evaluation

#### TS of *Ipomoea mauritiana* tuber

The *Ipomoea mauritiana* tuber's transverse section shows an outer layer of cork, followed by a cortex containing parenchyma cells packed with calcium oxalate rosette crystals and starch grains, and xylem tissue in the middle. Figure 2 shows the thin section of *Ipomoea mauritiana* tuber.

### Powder microscopy

Powder microscopy is the process of studying different plant structures like bordered pitted vessels, cork cells, non-lignified fibers, parenchyma cells, colored content, lignified fibers, tracheids, and starch grains to recognize and analyze the anatomical characteristics of plant material. Figure 3 shows the different identifying features of *Ipomoea mauritiana* tuber powder.

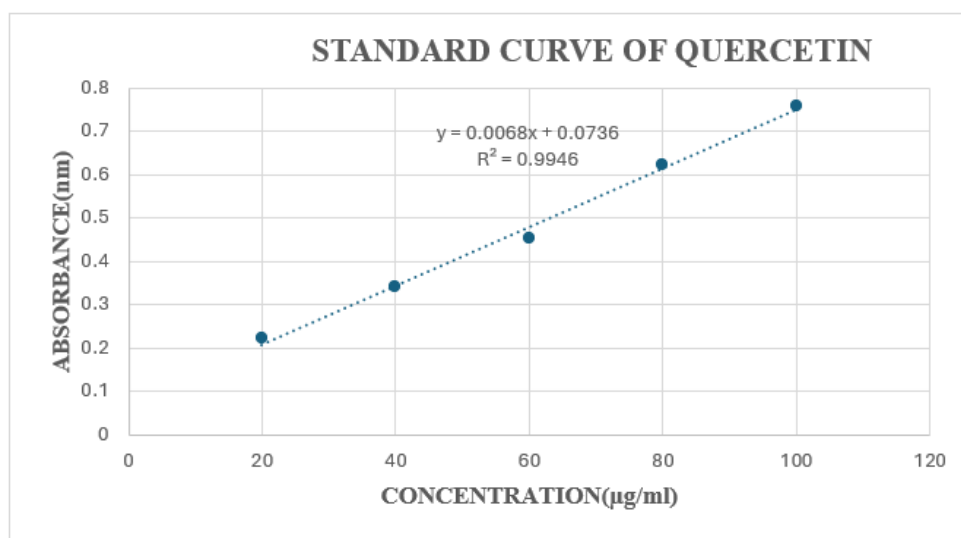


Figure 4: Standard curve of Quercetin.

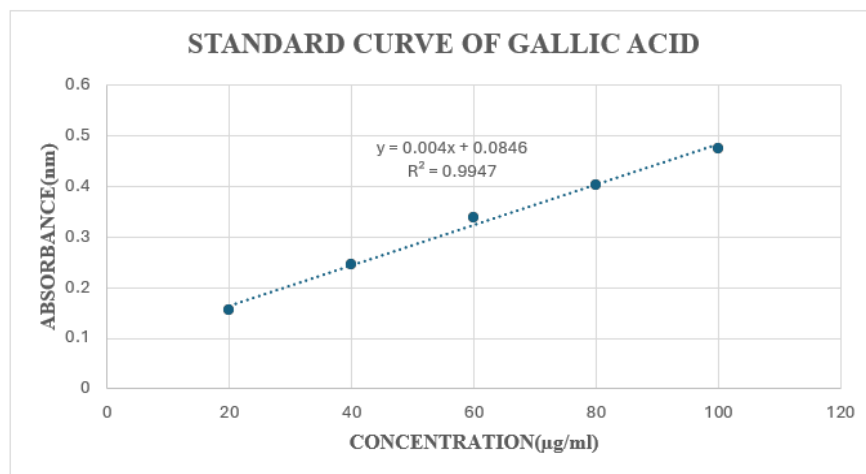


Figure 5: Standard curve of Gallic acid.

### Physicochemical evaluation

The result of physicochemical evaluation of *Ipomoea mauritiana* tuber is given in the Table 1. Physicochemical parameters, such as a low moisture content of 3.4%, suggest good stability against microbial growth, while the ethanol extractive value of 8.5% w/w highlights ethanol as an effective solvent for extracting polar bioactive compounds like flavonoids and phenolics. The high foaming index (>100) indicates a significant saponin presence, supporting its traditional use as a galactagogue and tonic.

### Phytochemical screening

The tuber of *Ipomoea mauritiana* contains high levels of carbohydrates, flavonoids, phenols, and terpenoids, with low levels of alkaloids, tannins and glycosides, and lacks proteins. The presence of carbohydrates, terpenoids, alkaloids, glycosides, tannins, and saponins, all of which could work together to produce synergistic pharmacological effects. Table 2 shows presence of constituents in ethanol.

### Quantitative estimation of phytoconstituents

#### Total Flavonoid Content (TFC)

Employing the aluminium chloride colorimetric technique with quercetin as the standard ( $y = 0.0068x + 0.0736$ ,  $R^2 = 0.9946$ ), the flavonoid content of ethanol extract of *Ipomoea mauritiana* tuber was determined to be 26.11 µg Quercetin Equivalent (QE) per mg of extract determined from the standard curve of quercetin Figure 4.

#### Total Phenolic Content (TPC)

Applying the Folin-Ciocalteu method with gallic acid as standard ( $y = 0.004x + 0.0846$ ,  $R^2 = 0.9947$ ), the phenolic content of ethanol extract was found to be 54.3 µg Gallic Acid Equivalent (GAE) per mg of extract identified from the standard curve of Gallic acid Figure 5.

### CONCLUSION

Pharmacognostical and phytochemical characterization of the *Ipomoea mauritiana* tuber offers an extensive scientific basis for its identification, authentication, and quality assessment. The work proves the occurrence of imperative bioactive molecules responsible for its ancient applications in Ayurvedic medicine, i.e., in Ayurvedic formulations such as Chyawanprash. The excessive ethanol extractive value along with the identification of essential secondary metabolites justifies the medicinal effectiveness of the plant. Additionally, the precise physicochemical parameters act as key quality control parameters, minimizing risks of adulteration and misidentification. Overall, this study confirms the traditional claims and provides baseline data required for future pharmacological and clinical research.

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### ABBREVIATIONS

w/w: Weight by weight; NSS: Nair Service Society; TS: Thin Section; gm: Grams; mL: Millilitre; g/L: Gram per litre;  $H_2SO_4$ : Sulphuric Acid; HCl: Hydrochloric acid; NaOH: Sodium Hydroxide; TFC: Total Flavonoid Content; TPC: Total Phenolic Content; QE: Quercetin equivalent; GAE: Gallic acid equivalent.

### CONFLICT OF INTEREST

None.

### FUNDING

None.

## ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Not applicable.

## HUMAN AND ANIMAL RIGHTS

No animals/humans were used for studies that are the basis of this research.

## CONSENT FOR PUBLICATION

Not applicable.

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