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Preliminary phytochemical and Pharmacognostic study of *Ipomoea quamoclit* leave extract

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Abstract
Objective: Preliminary Phytochemical and Pharmacognostic study of *Ipomoea quamoclit* Linn of leaf extract.
Methods: The leaf was collected, shade-dried, and made into powder. The powdered plant material was subjected to successive solvent extraction by the Soxhlet extraction method using methanol as solvent. The extracts were subjected to preliminary phytochemical screening in which chemical tests were carried out for the detection of various phytoconstituents. In the pharmacognostic study the macroscopical examination was documented using a Nikon D-5600 Digital camera as well as and in microscopical examination the Sample was preserved in fixative FAA for more than 48 hr. The preserved specimens were cut into thin transverse sections using a sharp blade and the sections were stained with 0.8% safranin and 0.5% astra blue. Transverse sections were photographed using an Axiolab 5 trinocular microscope attached to the Zeiss Axiocam 208 color digital camera under bright field light. Magnifications were indicated by the scale bar.
Conclusion: The present study reveals the preliminary phytochemical and Pharmacognostic study of *Ipomoea quamoclit* of leaf extract.

Keywords: *Ipomoea quamoclit* preliminary phytochemical and Pharmacognostic study

Introduction
Ipomoea quamoclit was originally an American plant and reached Europe by the 1550s. It is commonly known as the cypress vine; cardinal creeper is a species of vine in the family Covolvulaceae and the vines are recorded from both Europe and India in the 1500s and were taken to both places for their medical uses. It is a medicinal plant traditionally used to treat hemorrhoids, ulcers, diabetes, and cancer. According to Ayurveda, Cypress Vine plants are used to treat various ailments and are also an important ingredient in the preparation of some medicinal products. However, they are widely grown throughout the tropics as ornamental plants for their attractive flowers and exotic leaves. This ornamental plant grows as an annual plant that thrives only in tropical and subtropical regions. In non-tropical areas, these are grown as seasonal plants. *Ipomoea quamoclit* L is a less-studied medicinal plant that is used as folk medicine around the world for illness. The plant is considered cooling and purgative; used in chest pain, and pounded leaves are used as a remedy for bleeding piles and carbuncles. It belongs to the Convolvulaceae family and is an annual, herbaceous plant, commonly known as Mayil Manikkam, Akasamulla, Kunjalata, Tarulata, Kamalata, Getphul in India and distributed throughout the tropical areas of the world. It is one of the commonly cultivated members of the Convolvulaceae, and arguably the most strikingly beautiful morning glory in the horticultural trade. These scarlet flowered climbers form a delicate, lacy mass of pinnately divided leaves during the warm months in temperature regions, and year-round in tropical areas. Various studies have confirmed that *Ipomoea quamoclit* exhibits a vast range of bioactivities like antioxidant activity, antimicrobial activity, anticancer activity, antidiabetic activity as well and insecticidal activity.

Scientific classification
Domain: Eukaryota
Kingdom: Plantae
Phylum: Spermatophyta subphylum: Angiospermae Class: Dicotyledonae

Order: Solanales
Family: Convolvulaceae
Genus: Ipomoea
Species: *Ipomoea quamoclit*

Botanical Information
Scientific Name: *Ipomoea quamoclit* L
Common Name: Cypress vine

Methodology
Soxhlet extraction method

- 35 g dried *Ipomoea quamoclit* leaves powder was weighed into the thimble.
- 350 ml of methanol was used as a
- solvent and placed in the boiling flask before fixing the thimble into the soxhlet apparatus.
- This setup was heated for about 24hrs.
- The extract was distilled in a vacuum under pressure to remove the solvent completely.
- It was dried and kept in desiccators till
- experimentation. The obtained extract was weighed and % yield was calculated in terms of air-dried powdered crude material.

The yield and % yield of various extracts of powdered leaves of *Ipomoea quamoclit* were reported in Table No.1

Qualitative phytochemical
Analysis
Phytochemicals are natural chemical substances found in plants that have beneficial or bad health effects. The discovery of phytochemicals can be used to predict a plant's pharmacological efficacy. The Preliminary Phytochemical screening of plants is still popular, with Phytochemicals determined by qualitative tests.

Test for alkaloids
Dragendroff's test
A few ml filtrates added with 1-2 ml Dragendroff's reagents produce a reddish brown precipitate.

Mayer's test: A few ml of filtrate with 1-2 drops of Mayer's reagent in the side of the test tube produces a creamy white or yellow precipitate.

Wagner's test
A few ml filtrates with 1-2 drops of Wagner's test along with the side of the test tube to produce a brown or reddish precipitate.

Picric acid test
A few ml of filtrate with 3-4 drops of 2% picric acid solution to produce an orange color.

Tannic acid test
Acidified extract with 10% tannic acid solution to produce a buff color precipitate.

Test for carbohydrates
Resorcinol test
2 ml filtrate aqueous extract solution and few crystals of resorcinol and an equal volume of conc. HCL and Heat to produce a rose color.

Test for reducing sugar
Fehling's test
1 ml each of Fehling's solutions A & B add 1 ml filtrate and boil with a water bath to produce a red precipitate.

Test for cardio glycosides
Keller-kalian test
1 ml filtrate is added to 1.5 ml glacial acetic acid and 1 drop of 5% ferric chloride and concentrated H_2SO_4 along the side of the test tube to produce a blue-colored solution.

Bromine water test
Plant extract is added to a few ml of bromine water to produce a yellow precipitate.

Test for proteins and amino acid
Ninhydrin test
2 ml filtrate and add 2 drops of Ninhydrin solution to produce a purple-colored solution.

Biuret test
2 ml of filtrate is mixed with 1 drop of 2% copper sulfate solution and 1 1 ml of 95% ethanol and KOH pellets to produce a pink-colored solution.

Ninhydrin test
2 ml filtrates add 2 drops of Ninhydrin solution to produce a purple-colored solution.

Xanthoproteic test
Plant extract is added to nitric acid to produce a yellow color solution.

Test for flavonoids
Lead acetate test
1 ml plant extracts and add a few drops of 10% lead acetate solution to produce a yellow precipitate.

Ferric chloride test: Extract the aqueous solution and add a few drops of 10% ferric chloride solution to produce a green precipitate.

Ammonia test
The filtrate is added to 5 ml of dilute ammonia solution and conc. H_2SO_4 to produce a yellow color solution.

Conc. H_2SO_4 test
The plant extract is added to conc. H_2SO_4 to produce an orange color.

Test for phenolic compounds
Gelatin test
Plant extract is dissolved in 5 ml of distilled water and 1% of gelatin solution and add 10% NaCl to produce a white precipitate.

Ferric chloride test
Aqueous extract is added to a few drops of 5% ferric chloride solution to produce a dark green or bluish-black color.

Lead acetate test
Plant extract is dissolved in 5 ml of distilled water and 3 ml of 10% lead acetate solution.

Test for tannins
Braymer's test
1 ml filtrate added to 3 ml distilled water and 3 drops of 10% ferric chloride solution to produce blue blue-green color.

Lead subacetate test
1 ml filtrate and add 3 drops of lead sub acetate solution to produce a creamy gelatinous precipitate.

Bromine water test
10 ml of bromine water add 0.5 gm of plant extract decoloration of bromine.

10% NaOH test
1.4 ml of plant extract adds 4 ml of 10% NaOH and shakes well causing the formation of emulsion.

Test for phlobatannins
HCL test
2 ml aqueous extract with an ml 1% HCL boiled to produce a red precipitate.

Test for saponins
Foam test
1.5 gm plant extract add 2 ml of water and vigorously shake to produce persistent foam for 10min.

Haemolysis test
A drop of fresh blood on a glass slide and add plant extract to produce a zone of hemolysis.

Test for phytosterols
Salkowsi's test
A few ml of filtrate and add few drops of H_2SO_4 produce a red color.

Libermann-Burchard's test
Plant extract is dissolved in 2 ml acetic anhydride and adds 1-2 drops of conc. H_2SO_4 is along the side of the test tube to produce an array of color changes.

Hesse's response
5 ml aqueous extract add 2 ml of chloroform and add 2 ml of conc. H_2SO_4 pink or red color ring is produced in the lower layer of the chloroform layer.

Acetic anhydride test
0.5 ml plant extract add to 2 ml of acetic anhydride and 2 ml conc. H_2SO_4 changes color from violet to blue/green.

Test for cholesterol
2 ml extract with 2 2 ml chloroform and 10 drops of acetic anhydride and add 2-3 drops of conc. H_2SO_4 to produce red-rose color.

Test for terpenoids
2 ml of chloroform add 5 ml of plant extract evaporate in a water bath and add 3 ml conc. H_2SO_4 to produce a grey colored solution.

Test for triterpenoids
Salkowski's test
A few ml of filtrate and a few drops of conc. H_2SO_4 to produce a golden yellow layer.

Test for diterpenes
Copper acetate test
Plant extract is dissolved in distilled water and 2-4 drops of copper acetate solution to produce an emerald green color.

Test for quinones
Conc. HCL test
Plant extract is added to conc. HCL to produce a green color.

Sulphuric acid test
10mg extract is dissolved in isopropyl alcohol and a drop of conc. H_2SO_4 to produce a red color.

Test for anthraquinones
Borntrager's test
10 ml 10% ammonia solution is added to a few ml of filtrate shake vigorously for 30 seconds to produce a pink, violet, or red color solution. Ammonia hydroxide test 10 mg of extract is dissolved in isopropyl alcohol and drop conc. ammonium hydroxide solution, formation red color after a few minutes.

Test for anthocyanins
HCL test
2 ml of plant extract is added to 2 ml of 2N HCL to produce a pink-red solution which turns blue violet after the addition of ammonia.

Test for carboxylic acid
Effervescence test
1 ml plant extract is added to 1 ml sodium bicarbonate solution appearance of effervescence.

Test for coumarins
NaOH test
Plant extract is dissolved in 10% NaOH and chloroform to produce a yellow color.

Test for resin: Acetic anhydride

1 ml plant extract is added to acetic anhydride solution and 11 ml conc. H_2SO_4 to produce orange to yellow.

Turbidity test

10 ml extract is added to 20 ml 4% HCL to produce turbidity. Phytoconstituents of various extracts of powdered leaves of *Ipomoea quamoclit* were reported in Table No. 2

Results and Discussions

The leaf of this plant was extracted by methanol as a solvent by using the Soxhlet apparatus. The semisolid extract is obtained as.

Table 1: Extractives values of methanol extract of powdered leaves of *Ipomoea quamoclit*

S.no	Solvent	Percentage yield
1	Methanol	5.569 g

Table 2: Phytoconstituent of methanol extract of powdered leaves of *Ipomoea quamoclit.*

Acetic anhydrideS.no	Phytoconstituents	Methanol Extract
1	Alkaloids	+
2	Carbohydrates	+
3	Reducing Sugar	-
4	Cardiac Glycosides	+
5	Protein and amino acid	+
6	Flavonoids	+
7	Phenolic Compounds	+
8	Tannins	+
9	Plobatannins	-
10	Saponins	+
11	Phytosterols	-
12	Cholesterols	-
13	Terpenoids	+
14	Triterpenoids	-
15	Diterpenes	-
16	Quinones	-
17	Anthraquinones	-
18	Anthocyanins	-
19	Carboxylic Acid	+
20	Coumarins	+
21	Resins	-

Note: Presence (+) Absence (-)

Pharmacognostic studies material and methods for anatomical studies

Collection of Specimens

The plant specimens for the proposed study were collected from Siddha Medicinal Garden, Mettur. Care was taken to select healthy plants and normal organs. The required samples of different organs were cut and removed from the plants and fixed in FAA (Formaline-5 ml + 70% Ethyl alcohol-90 ml). After 24 hours of fixing, the specimens were dehydrated with a graded series of tertiary-butyl alcohol as per the schedule by Sass, 1940. Infiltration of the specimens was carried by gradual addition of paraffin wax (melting point 58-60 C) until TBA solution attained supersaturation. The specimens were cast into paraffin blocks.

Sectioning

The paraffin-embedded specimens were sectioned with the help of a Rotary Microtome. The thickness of the section was 10-12 µm. dewaxing of the sections was by customary procedure (Johansen, 1940). The section was stained with Toluidine blue as per the method published by O'Brien *et al*. (1964). Since Toluidine blue is a polychromatic stain. The staining results were remarkably good, and some cytochemical reactions were also obtained. The dye rendered pink color to the cellulose walls, blue to the lignified cells, dark green to suberin, violet to the mucilage, blue to the protein bodies, etc. wherever necessary sections were also stained with safranin and fast-green and IKI (for starch).

For studying the stomata morphology, venation pattern, and trichome distribution, peridermal sections (sections taken parallel to the surface of the leaf) as well as clearing of the leaf with 5% sodium hydroxide or epidermal peeling by partial maceration employing Jeffrey's maceration fluid (Sass, 1940) were prepared. Glycerine- mounted temporary preparations were made for macerated/cleared materials. Powdered materials of different parts were cleared with NaOH and mounted in a glycerine medium after staining. Different cell components were studied and measured.

Photomicrographs

Microscopic descriptions of tissues are supplemented with micrographs wherever necessary. Transverse sections were photographed using an Axiolab 5 trinocular microscope attached to the Zeiss Axiocam 208 color digital camera under bright field light. Magnifications were indicated by the scale bar.

Macroscopy report

Leaves

Leaves are green colored, pinnately compound with 15 to 18 leaflets, imparipinnate, leaflets simple, sessile, linear, margin entire, apex acute, measuring 1.5 to 2 cm length and 0.2 to 0.4 cm breadth; petiole is 2 to 3 cm long (Fig.1).

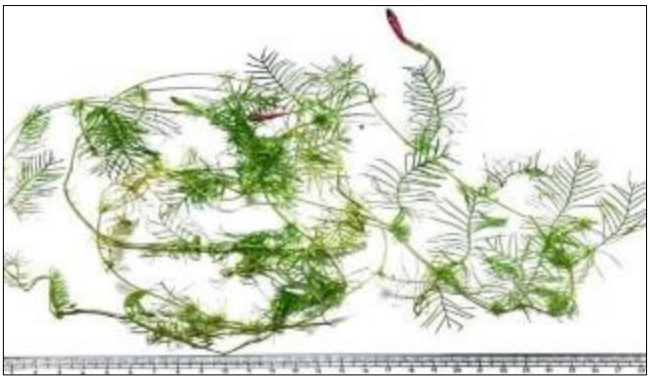


Fig 1: Macroscopy of Ipomoea quamoclit

Microscopy report

Petiole

TS of the petiole is irregularly shaped with a concave upper surface having wing-like projections and a deeply wavy lower surface; the epidermis is single-layered and covered by thin cuticles followed by 10 to 12 layers of cortical parenchyma cells; few oil globules and brownish mass contents are present in parenchyma; conjoint, collateral vascular bundles present at the center in which ecrescent-

shaped xylem is surrounded by phloem; two small trace bundles can be seen below the wing region in the cortex; pith is absent (Fig. 2).

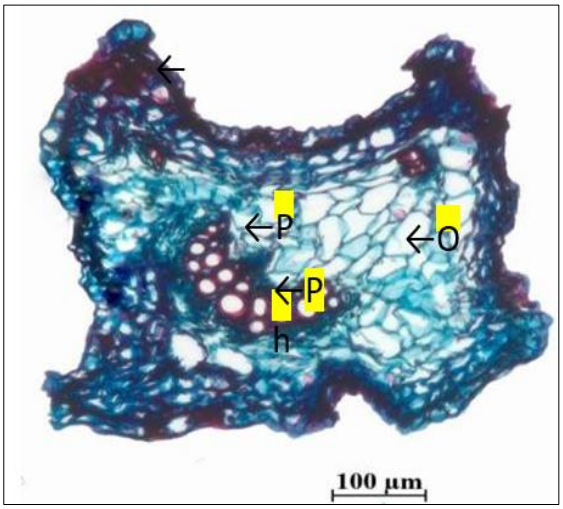
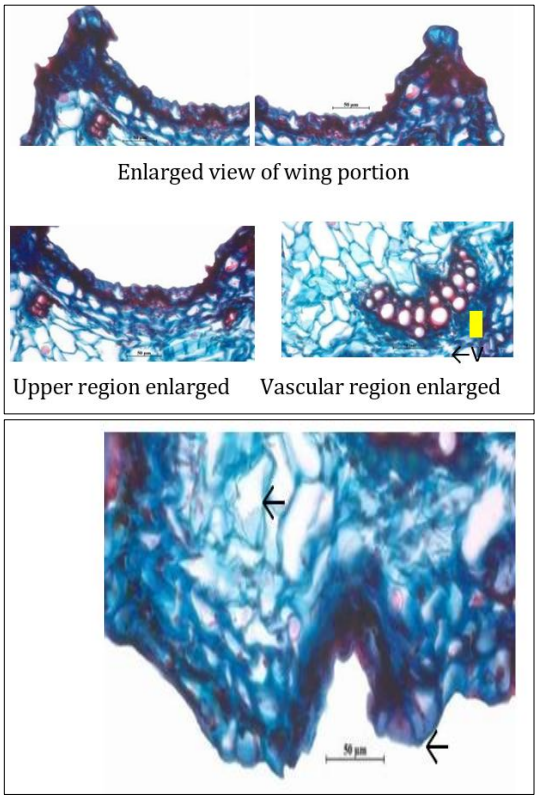


Fig 2: TS of *Ipomoea quamoclit* petiole TS of petiole



E - Epidermis; OG - oil globule; Pa - parenchyma; Ph - phloem; TB - trace bundle;V- vascular bundle LEAF

Fig 3: TS of the leaf shows an elevated upper midrib and wavy lower midrib with lateral laminar extensions.

Midrib

TS of leaf passing through midrib shows upper and lower epidermis, with few covering trichomes; beneath the epidermis some layers of cortical parenchyma are present continuous with central conjoint, collateral vascular bundle; xylem is found facing towards lower side and phloem on the upper side; mucilage cavity is present in ground tissue (Fig. 3).

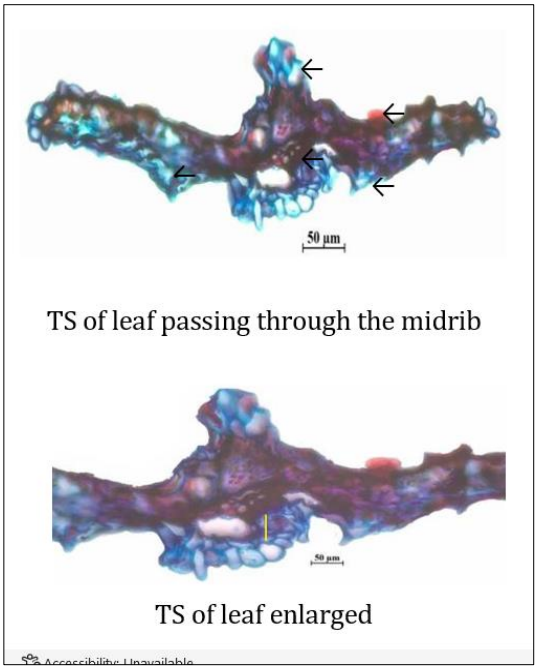
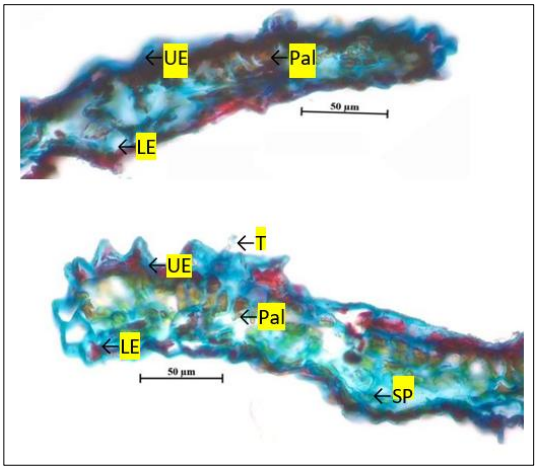


Fig 3: TS of *Ipomoea quamoclit* leaf

Lamina

TS of lamina shows upper and lower epidermis covered by thin cuticle and bears few covering and glandular trichomes followed by mesophyll tissue; mesophyll tissue is differentiated into a layer of palisade cells followed by 2 to 3 layers of loose lyarranged spongy parenchymatous cells; few oil globules are found in mesophyll cells (Fig. 3).



Cu - cuticle; GT - glandular trichome; LE - lower epidermis; MC - mucilage cavity; Mes - mesophyll tissue; Pal - palisade cells; Ph - phloem; SP - spongy parenchyma; T - trichome; UE - upper epidermis; VB - vascular bundle; Xy – xylem

Fig 4: TS of lamina

Conclusion

This present study provides information on the phytochemical evaluation and pharmacognostic studies of *Ipomoea quamoclit*. For chemical analysis, the leaf of *Ipomoea quamoclit* was chosen. The qualitative study of the *Ipomoea quamoclit* leaf sample yielded favorable results for a variety of chemical components. The extract from the solvents was examined

for a variety of compounds using established procedures, yielding the following results. The methanol extracts of the product produce 5.569g and Alkaloids, Flavanoids, Tannins, Coumarins, Carboxylic acid, Terpenoids, Saponins, Carbohydrates, Phenolic compound, Cardiac Glycosides, Protein and amino acid chemical compounds was presented. The study's a findings can be useful source of knowledge and give appropriate criteria for identifying this plant material in future research and applications.

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