Pharmacognostic, Physicochemical, and Phytochemical Evaluation of *Ipomoea reniformis* Choisy: An Ethnomedicinal Plant Native from Maharashtra, India

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ABSTRACT

Background: The genus Ipomoea consists of many climbing herbs and weeds belonging to the family Convolvulceae which are distributed all over India and are extensively important in Ayurvedic, Unani, and Siddha medicine. Objectives: Looking into its above important activity the present research studies were carried out to investigate the Ipomoea reniformis Choisy ethnomedicinal plant for its quality, purity, and identification with the help of Pharmacognostic, Physicochemical, and Phytochemicals evaluation. Materials and Methods: The selected and authenticated medicinally important plant *I. reniformis* is belonging to the family Convolvulceae with the help of Pharmacognostic and Physicochemical characterization of powder and Phytochemicals evaluation of various extracts for its quality, purity, and identification. Results: Macroscopic studies of the plant part's color, odor, size, taste, and arrangement of leaves, Petiole, and stem of I. reniformis were observed. Microscopy of leaves, petiole, and stem of the I. reniformis plant confirmed the presence of several significant sections. Physiochemical parameters such as % moisture content, % total solid content, total Ash values, water and alcohol soluble ash, acid-insoluble ash, and Sulfated ash values (% w/w) are 1.15±0.02, 98.85±0.04, 4.91±0.14, 3.28±0.27, 4.02±0.22, 0.93±0.37, 1.31±0.35 respectively. Ethanol and methanol produced high extractive yields of 11.06% and 9.86% respectively. Phytochemical evaluation of the various herbal extracts found many significant phytochemical constituents in ethanol and methanol extracts. Conclusion: Pharmacognostic, physiochemical, and phytochemical studies were carried out for the identification and authentication of *I. reniformis* Choisy, and these studies will help in the differentiation of the medicinal plants from its various species.

Keywords: Fluorescence Analysis, *I. reniformis*, Percent yield, Pharmacognostic studies, Physicochemical, Phytochemicals evaluation.

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INTRODUCTION

Many of the population throughout the world have returned to the utilization of medicinal plant products in the healthcare system in recent years and also in many countries, conventional medicine forms a fundamental part of the healthcare system. Medicinal plants are useful for both reducing and treatment of human diseases with the help of major Phytochemicals constituents. The increasing demand for replacement medicine has resulted in the use of natural plant treatment producing a



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great need for knowledge on the properties and uses of medicinal plants; resulting in the evolution of natural product markets and importance to the conventional medicinal systems.^[1-3] With more than 500 species, the largest genus in the Convolvulaceae family is Ipomoea. The world's tropical and subtropical regions provide habitat to species of the Ipomoea genus. Ipomoea is used by people because it contains many therapeutic and psychotropic substances, primarily alkaloids. Food crops of the genus include sweet potatoes and water spinach leaves, which have significant commercial importance.^[4] In India, about 65% of the people depend on ethnomedicine for their major healthcare requirements.^[5,6] *Ipomoea reniformis* (I.R) has another name *Merremia emarginata* (Burm.f.) Hallier f. is a horizontally growing herb related to the Convolvulceae family (Figure 1). It is a much-branched herb, growing several years on ground places

and having therapeutic use. Found in the farm, damp places. It is regularly found in the Indian subcontinent, Malaysia Sri Lanka, Tropical Africa, and the Philippines. In India, it is regularly found in Tamil Nadu, Karnataka, Maharashtra, Kerala, Bihar, Gujarat, and Rajasthan rising to 900 m in the hills.^[7-9]

Looking into its above important activity the present research studies were carried out to investigate the *I. reniformis* (Choisy) ethnomedicinal plant for its quality, purity, and identification with the help of Pharmacognostic, Physicochemical, and Phytochemicals evaluation.

MATERIALS AND METHODS

Selection and Authentification of the Medicinal Plant

The whole plant of *I. reniformis was* collected from the local farm of Aurangabad district between July to December, and it was identified and authenticated as having Accession no. Bot./2020/0720 by Botany Department, Dr. Babasaheb Ambedkar Marathwada University, Aurangabad, Maharashtra, India.

Chemicals and Instruments

All the chemicals used in this study were analytical grade obtained from Fine-Chem Chem. Ltd., (Mumbai, India) and SRL Pvt. Ltd., (Mumbai, India). Scanning Electron Microscopy (SEM), digital camera and other common glassware and instruments, and staining reagents were used for the study.

The evaluations of the macroscopic, microscopic, Fluorescence, physicochemical, % percentage yield of various extracts, and phytochemical parameters were determined according to standard methods.

Pharmacognostic study

Macroscopic Studies

The macroscopic evaluation of fresh aerial parts of *I. reniformis* such as color, appearance, size, shape, and arrangement of leaves, stems, and petiole of plants was observed.^[10-12]

Microscopic Studies

A microscopic study was carried out on the *I. reniformis* plant by preparing thin sections of leaves, Petiol, and stem. Safranin was used twice to stain the thin portions, and quick green was then rinsed with water. Dehydrating these parts that have been double-stained for one minute at a time with alcohol of increasing strength (30%, 50%, 70%, 90%, and finally 100%), and placed in glycerine for inspection and lignification confirmation (10x, 40x). SEM has been used to image plants at high resolution. Photographs at different magnifications were taken by using a digital camera.^[13-16]

The drying process of the medicinal plant material

The shade drying method is the most acceptable form of drying which involves less exposure to heat and there are fewer chances of chemical alteration. According to these methods, the whole plant parts were dried using air undergoing shade conditions at normal room temperature (20-22°C) for 15 days. The dehydrated leaves pass from sieves no. 20 and 22 to become coarse powder stored at 4°C in an airtight bottle for further studies.^[17]

Physicochemical analysis

Moisture content (loss on drying), total solid content, Ash values in the powder, acid-insoluble ash, Sulfated ash, water-soluble extractives, and alcohol-soluble extractives of *I. reniformis* plant powder determined using standard literature procedure.^[18-25]

Fluorescence Analysis

Powdered Ariel Part material was treated with a variety of chemical and inorganic reagents before being examined under visible and long UV light like 50% HCl, 50% H_2SO_4 , 50% HNO₃, and 10% NaoH, Petroleum ether, chloroform, Ethanol, Ethylacetate and HCl (1:1).^[26]

Preparation of plant extracts by Soxhlet extraction using various solvents

The following solvents are used for the extraction of Plant material by the Soxhlet extraction process followed by standard literature methods (Table 1).

Extraction Plant material

Dry the aerial plant material using Shade drying method and passes from sieve No. 20, 22 for making coarse material of medicinal plant (1.2 kg of each plant respectively). The powder of plant material was extracted with 95% ethanol (60°C) using Soxhlet extraction apparatus. Each batch of material contains 180 g coarse powder. Filter each time and remove the solvent from the combined extract under dried by vaccum Rota evaporator to get 92.21 g of extract.^[27-30]

Nature, color, and extractive yield of various extract

Estimation of the nature, color, and extractive value of 6 solvent extracts such as Aqueous, Methanol, Ethanol, Chloroform, Petroleum ether, and Acetone was done according to the standard literature method.

Preliminary phytochemicals evaluation

Preliminary Phytochemicals evaluation is the important step used after extraction to identify different groups of bioactive components present in plants of *I. reniformis* extract. Aqueous, Methanol, Ethanol, Chloroform, Petroleum ether, and Acetone extracts were analyzed for identification test of the presence or absence of various phytochemicals by using the standard Procedure mentioned in the literature.^[31-37]

RESULTS

Pharmacognostic Studies

Macroscopic studies of I. reniformis

It has always been possible to quickly and accurately identify plant material using the macroscopic character, which also acts as one of the essential standards (Table 2).

Microscopic studies of *I. reniformis T.S of the leaf*

Leaf lamina was dorsiventral. The upper epidermis was single-layered, covered with a single layer of cuticle with anomocytic stomata. Trichomes are absent. Spongy parenchymas have 3 to 4 layers with thin walls of the cells. Some cells contain calcium oxalate. Palisade cells with a single layer are present. The lower epidermis was similar to the upper epidermis single-layered, covered with a single layer of cuticle with anomocytic stomata. Trichomes are absent. Midrib with a biconvex outline, a single layer of cuticle covering the upper and lower epidermis. Collenchyma present below the upper and lower epidermis having 3 to 4 layers of thick-walled cellular parenchyma. Lignified xylem and phloem bowl-shaped vascular bundles are present with some starch grains (Figure 2).

T.S. of the Petiole

The upper epidermis was double-layered with rectangular cells and semi-circular towards the laminal side. Trichomes are present. 5 to 6 layers of Spongy parenchyma. Thin-walled parenchymatous cells with pith present. Some cells contain calcium oxalate. Palisade cells with double layers are present. The lower epidermis was similar to the upper epidermis double-layered with rectangular cells shape and semi-circular in shape towards the laminal side. Trichomes are present. Lignified 5 to 6 xylem and phloem vascular bundles are present with some starch grains (Figure 3).

T.S. of the stem

It was cylindrical, the cortex consisted of 4-5 layers, and vascular bundles were surrounded by polygonal lignified parenchyma and Collenchyma cells. The epidermis was single-layered, quadrangular cells, thick-walled with anomocytic stomata and this epidermis was surrounded by cuticles.

Vascular bundles consisted of about 7-8 collaterals, open, and arranged in the ring structure. Contains well-developed xylem, and phloem containing sieve tubes. Pericyclic fibers- lignified with green color present on the upper side of the vascular bundle. The Pith was large and consisted of thin-walled cells consisting of polygonal parenchymatous cells. Fibers are in a group of lignified 2 to 3 cells (Figure 4).

Physico-chemical parameters of *Ipomoea reniformis* (Choisy) plants powder

The obtained results of the Physico-chemical parameters of *Ipomoea reniformis* (Choisy) plant powder (Table 3).

Fluorescence analysis of I. reniformis plants powder

Fluorescence characteristics of *I. reniformis* plant powder were observed in visible, short, and long UV light (Table 4).

Nature, color, and Percentage of extractive yield-

The phytochemicals are important bio-active compounds in nature, color, and qualitatively analysis of various extracts of *I. reniformis* like Aqueous, Methanol, Ethanol, Chloroform, Petroleum ether, and Acetone having different polarity of



Figure 1: Ipomoea reniformis (Choisy), Convolvulceae.

Table 1: Solvent used for extraction with Polarity, Boiling point, and Water Solubility.

SI. No.	Solvents used for the extraction process	Polarity	Boiling Point in °C	Water solubility (% w/w)
1	Aqueous (Hot-water)	9	100	100
2	Methanol	5.1	65	100
3	Ethanol	5.2	78	100
4	Chloroform	4.1	61.2	0.815
5	Petroleum ether	0.1	20-80	0.021
6	Acetone	5.4	56	100

SI. No.	Characteristics	Ipomoea reniformis (Choisy)		
1	Part of plant	Leaves	Stem	
2	Arrangement	Alternative	NA	
3	Size	0.5 to 3.5 cm	Up to 70 to 75 cm	
4	Shape	Kidney shape or ovate	Cylindrical in shape	
4	Color	Greenish	Greenish	
5	Appearance	Smooth	Smooth	
6	Petiole	2.5 to 4.5 cm long	NA	

Table 2: Macroscopic studies of *I. reniformis* leaves, stem, and petiole.

Table 3: Physico-chemical parameters of *Ipomoea reniformis* (Choisy) plants powder

Parameters	Values (% w/w)*
% Moisture content in powder	1.15±0.02
% Total solid content	98.85±0.04
Total Ash values in the powder	4.91±0.14
Water soluble Ash	3.28±0.27
Alcohol soluble Ash	4.02±0.22
Acid-insoluble ash	0.93±0.37
Sulfated ash	1.31±0.35

*Values express in mean±SD. SD: Standard deviation.







Figure 3: T.S of petiole.

Table 4: Fluorescence analysis of I. reniformis plants powder.

Reagents	Fluorescence analysis of powder			
	Observed under sunlight	Observed under UV light		
50% HCl	Brown	Yellowish -green		
$50\% H_2SO_4$	Brown	Bluish-green		
50% HNO ₃	Dark-yellow	Green		
10% NaOH	Brown	Greenish-yellow		
Ethanol	Dark-green	Yellowish-green		
Methanol	Dark-green	Yellowish-green		
Chloroform	Greenish-brown	Pale-green		
Petroleum ether	Light-green	Pale-green		
Acetone	Greenish-brown	Dark brown		
Aqueous	Light green	Dark-green		

chemicals results in % Yield of extracts by maceration process was calculated in gram and converted it into % w/w in Table 5.

Phytochemical evaluation

Preliminary and specific Phytochemicals studies prove that various bio-active Phytochemicals are present in *I. reniformis* (Table 6).

DISCUSSION

On the basis of the literature survey *I. batatas, I. digitata*, I *stans, I. turpethum* commonly occurring medicinal plants belonging to the genus Ipomoea. The family Convolvulaceae has 20 genera and 158 species in India, the majority of which are found in the country's south and west. It has over 60 known species of *Ipomoea* in India and 24 in Maharashtra state. These species are quickly becoming extinct from a variety of locations since people are unaware of the benefits of the plant.^[38] They have many important chemical constituents and also had claimed to cure many diseases according to recent studies. The Pharmacognostic, Physicochemical, and Phytochemical investigations are primary steps for identification and standardization of Ethnomedicinal herbs has increased rapidly in recent years medicinal plants remain more reliable

SI. No.	Solvent used	Nature	Color	Yield of extract	
				gram	% w/w
1	Aqueous	Amorphous	Light green	2.13	7.12
2	Ethanol	Sticky	Dark green	3.48	11.06
3	Methanol	Sticky	Dark green	2.96	9.86
4	Chloroform	Smooth	Greenish brown	1.07	3.56
5	Petroleum ether	Smooth	Light green	0.58	1.93
6	Acetone	Smooth	Greenish brown	1.68	5.60

Table 5: Nature, color, and % extractive yield of *I. reniformis* extract.

Table 6: Qualitative analysis of Phytochemicals of I. reniformis extracts.

Phytochemicals Analysis	Methanol	Ethanol	Chloroform	Petroleum Ether	Acetone	Hot water
Alkaloids						
Glycosides	+++	+++	+++		+++	+++
Flavonoids	+++	+++	+++	++	+++	+++
Coumarin glycosides	+++	+++			+++	++
Anthraquinone Glycosides	++	++			++	
Cardiac Glycosides	++	++	++		++	++
Tannins	+++	+++	++	++	+++	++
Steroids	++	++	++		++	++
Saponins	++	++			++	++
Resins	++	++			++	++
Terpenoids	++	++	++		++	++
Carbohydrates	+++	+++	+++	++	+++	+++
Proteins	+++	+++			+++	+++
Amino acids	++	++		++	++	++

(Present Phytochemicals indicating +, Absent Phytochemicals indicating -).



Figure 4: T.S of stem.

than despite the availability of modern technologies. It will useful in the authentification of Ethnomedicinal plants.^[39]

The macroscopic evaluation revealed specialize features like color, appearance, size, shape, and arrangement of leaves, stems, and petiole of fresh aerial parts of *Ipomoea reniformis* (Choisy) it can serve as diagnostic parameters for differentiate with others similar plant species (Table 2).

The microscopic evaluation of *Ipomoea reniformis* leaf shows the upper epidermis was with anomocytic stomata. Presence of Spongy parenchyma, calcium oxalate, Palisade cells, collenchyma, thick-walled cellular parenchyma, lignified xylem and phloem bowl-shaped vascular bundles are present with some starch grains. Trichomes are absent (Figure 2).

T.S. of the Petiole shows the upper epidermis was double-layered with rectangular cells shape are present. Presence of Trichomes, Spongy parenchyma. Thin-walled parenchymas, calcium oxalate, Palisade cells, lignified 5 to 6 xylem and phloem vascular bundles are present with some starch grains (Figure 3). T.S. of the stem shows cylindrical shape with cortex, vascular bundles were surrounded by polygonal lignified parenchyma and Collenchyma cells. The epidermis was single-layered, quadrangular cells, thick-walled with anomocytic stomata and this epidermis was surrounded by cuticles. Pericyclic fibers present on the upper side of the vascular bundle. The Pith was large, thin-walled of polygonal parenchymatous cells. Fibers also present (Figure 4). Additionally, this research might contribute to reducing the potential of adulteration and provides quality of Ethnomedicinal plants.

Physicochemical evaluation includes Moisture content $(1.15\pm0.02\% \text{ w/w})$ has not more than 14% w/w thus inhibited bacterial, fungal and yeast growth, total solid content (98.85±0.04% w/w), total Ash values (4.91±0.14% w/w) in the powder significantly important for purity, quality and standardization of herbal drugs and also useful in identification of foreign inorganic matter, acid-insoluble ash(0.93±0.37% w/w), sulphated ash (1.31±0.35% w/w), water-soluble extractives (3.28±0.27% w/w), alcohol-soluble extractives (4.02±0.22% w/w) of *Ipomoea reniformis* (Choisy) plant powder was evaluated (Table 3).

Fluorescence analysis of *I. reniformis* plants powder reacted with reagent 50% HCl, 50% H_2SO_4 , 50% HNO_3 , 10% NaoH, Ethanol, Methanol, Chloroform, Petroleum ether, Acetone, Aqueous. It has shown varies significant color observed under UV light and Sunlight (Table 4).

The Solvent used for *I. reniformis* extraction with their Polarity, Boiling point, and Water Solubility (Table 1). Maximum extractive yield found in extracts of Ethanol (11.06%), Methanol (9.86%) and Aqueous (7.12%) as compared to extracts of Petroleum ether (1.93%), Chloroform (3.56%), and Acetone (5.60%) (Table 5). So, solvent ethanol selected for Soxhlet extraction because of providing high yield capacity of extract, less toxic than other methanol and other solvents, less time required for extraction and available cheapest cost.

Ipomoea reniformis ethanolic and methanolic extracts was found many valuable phytochemicals contain glycosides, flavonoids, coumarin glycosides, anthaquinone glycosides, cardiac glycosides, tannins, steroids, saponins, resins, terpenoids, carbohydrates, proteins, amino acids. This phytochemicals can serve in treatment of various diseases (Table 6).

CONCLUSION

Pharmacognostic and phytochemical studies were carried out for the identification and authentication of *Ipomoea reniformis* (Choisy). From the phytochemical studies, many presences of various phytochemicals are present in *Ipomoea reniformis* (Choisy). These studies will help in the differentiation of the medication from its various species. It is necessary for the correct identification of the plant and can serve for future scientific findings as less information was found reported during the exhaustive literature survey. This research would contribute to the differentiation of the medication from its various species.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest. The authors alone are responsible for the content of the paper.

ABBREVIATIONS

I.R: *Ipomoea reniformis* (Choisy); T.S: Transverse section; SEM:
Scanning electron microscopy; SP: Spongy Parenchyma; X:
Xylem; Ph: Phloem; UE: Upper Epidermis; LE: Lower Epidermis;
P: Palisade; C: Collenchyma; Ep: Epidermis; La: Lamina; MR:
Midrib; AS: Anomocytic Stomata; Pf: Pericyclic fibers; T:
Trichomes; Pt: Pith; Sg: Starch grains; Pp: Polygonal parenchyma;
Cr: Cortex.

SUMMARY

The genus Ipomoea belonging to the family Convolvulceae which are distributed all over India and are extensively important in Ayurvedic, Unani, and Siddha medicine. In the present research studies were carried out to investigate the Ipomoea reniformis Choisy ethnomedicinal plant for its quality, purity, and identification with the help of Pharmacognostic, Physicochemical, and Phytochemicals evaluation. Macroscopic studies of the plant part's color, odor, size, taste, and arrangement of leaves, Petiole, and stem of I. reniformis were observed. Microscopy of leaves, petiole, and stem of the I. reniformis plant confirmed the presence of several significant sections. Physiochemical parameters such as % moisture content, % total solid content, total Ash values, water and alcohol soluble ash, acid-insoluble ash, and Sulfated ash value was evaluated. Ethanol and methanol produced high extractive yields. Phytochemical evaluation of the various herbal extracts found many significant phytochemical constituents. Pharmacognostic, physiochemical, and phytochemical studies were carried out for the identification and authentication of I. reniformis Choisy. It can serve for future scientific findings as less information was found reported during the exhaustive literature survey. This research would contribute to the differentiation of the medication from its various species.

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