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Antioxidant and antibacterial activity of *Ipomoea mauritiana* Jacq.: a traditionally used medicinal plant in Bangladesh

Istiaq Alam¹, Shaekh Forid^{1,2}, Miah Roney², Fadhilzil Fasihi Mohd Aluwi² and Moyeenul Huq^{1,3*}

Abstract

Background: In an attempt to explore the scientific basis for the pharmacological benefits the antioxidant and antimicrobial activities of *Ipomoea mauritiana* whole plant methanol extract were assessed.

Methods: The total phenolic and flavonoid content were determined using standard method while antioxidant activity was determined by DPPH free radical scavenging activity method. The antimicrobial activity was evaluated by disk diffusion method and compared with standard kanamycin (30 µg/disc).

Results: The results revealed that *I. mauritiana* extract contains tannin, saponin, terpenoids, alkaloid and flavonoids. In DPPH, ascorbic acid and extract showed highest scavenging activity and it was 90.96% at concentration 800 µg/mL and *I. mauritiana* methanol extract showed 72.28% at a concentration of 800 µg/mL. The extract was able to reduce the stable free radical DPPH with an IC₅₀ of 275.084 µg/mL while that of ascorbic acid was 230.09 µg/mL. Total phenolic constituent of the extract was 59.302 ± 3.289 mg/g as gallic acid equivalent. The flavonoid content of methanolic extract of *I. mauritiana* was 27.212 mg of QE/g. In case of antimicrobial screening, crude extracts of *I. mauritiana* showed notable antibacterial activity against tested microorganisms. The extract and standard showed the highest mean zone of inhibition ranging from 13 to 19 mm and 37 to 42 mm, respectively at a concentration of 400 µg /disc and against the gram positive bacteria (*Bacillus cereus*-19.25 mm) showed highest zone of inhibition.

Conclusions: The results indicate that *I. mauritiana* possesses considerable antioxidant and antimicrobial activity.

Keywords: *Ipomoea mauritiana*, Antioxidant activity, Antimicrobial activity

Introduction

Ipomoea mauritiana (*I. mauritiana*) Jacq. (Synonyms: *Ipomoea digitata*; *Ipomoea eriosperma*; *Ipomoea paniculata*; *Convolvulus paniculatus*) is a vine of the Convolvulaceae family. The plant has ethnomedicinal importance and found in many parts of the world. In Ayurvedic industries, *I. mauritiana* is popularly used as “Vidari” instead of the permitted raw drug *Pueraria tuberosa* (Roxb. ex Willd). The Ayurvedic Pharmacopoeia of India

correlates *I. mauritiana* as Kshiravidari. Tubers are used in almost 45 Ayurvedic formulations, and in many instances also used as single drug. This is also an important component of the popular ayurvedic formulation Chyavanaprasha [1]. Traditionally the tubers are also extensively used by local communities in different parts of Bangladesh to treat various ailments including pain in spinal cord, to increase breast milk, body strength and sperm count, to control tuberculosis, as blood purifier in jaundice and to treat biliary disorders [2–5]. The leaves are also used to treat leucorrhoea and diabetes with obesity and in infrequent urination by the folk medicine practitioners of Kurigram and Jhalokati districts of Bangladesh [6].

* Correspondence: moyeenul.rph@uap-bd.edu

¹School of Science and Engineering, Department of Pharmacy, Southeast University, Dhaka, Bangladesh

³School of Medicine, Department of Pharmacy, University of Asia Pacific, 74/A, Green Road, Dhaka, Bangladesh

Full list of author information is available at the end of the article



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The pharmacological investigations reported analgesic, hypoglycemic, hypocholesterolemic, and hepatoprotective activities of *I. mauritiana* tuber root [7, 8]. Additionally, the antibacterial activity against *Escherichia coli* which caused catheter associated urinary tract infections has also been reported by Pavan [9]. A number of phytochemical have been isolated from *I. mauritiana* tuber root methanol extracts which includes taraxerol, taraxerol acetate, β -sitosterol, scopoletin and 7-O- β -D-glycopyranosylscopoletin (scopolin) [10]. Literature showed that, scopoletin isolated from *Lasianthus lucidus* possessed antibacterial activity against *Pseudomonas aerigenosa* [11]. Plants are major source of natural antioxidants that can serve as possible drug candidates for different chronic ailments such as neurological diseases, swelling and diabetes [12, 13]. The wide range of potential benefits of natural antioxidants has increased the curiosity largely in medicine to ascertain new molecules to combat oxidative stress caused by free radicals [13].

Infective microbes are one of the major public health threats causing morbidity and mortality worldwide [14]. Several life-threatening microorganisms have now become resistant to many commercial antibiotics [15]. Novel, safe, potent and wide-spectrum antimicrobial molecules are therefore urgently needed [16]. Even though the plant has been widely used as traditional medicine, however there is inadequate scientific evidence on the potential benefits of the whole plant. Therefore, the current study was designed to determine the antioxidant and antimicrobial activities of *I. mauritiana* whole plant methanol extract.

Materials and methods

Plant collection

I. mauritiana was collected from Tangail district of Bangladesh in July 2018 and was identified by Bangladesh National Herbarium. The collected plants (as a whole) were cleaned using tap water to remove the soil and washed with distilled water followed by shade drying. The dried plants were grounded into a fine powder.

Plant extract preparation

Powdered whole plant material having a weight of about 50 g were taken in one amber glass container and soaked in 500 mL methanol, for 3 days with occasional shaking and stirring. The extractive was then filtered using Whatman filter paper number 1 to attain a clear filtrate. The filtrate extract was concentrated and dried in vacuum at 40 °C and stored at 4 °C until further use.

Preliminary phytochemical screening

The presence of different phytochemicals such as alkaloid, steroids, glycosides, flavonoid, tannins, saponin, and terpenoids in crude methanol extract was determined following standard methods [17, 18]. Any color change or precipitation formation indicated positive reactions to these tests.

Test for steroids and Triterpenoids

In the extract solution, few drops of acetic anhydride added and boiled. After cooling, concentrated sulphuric acid was slowly poured into the test tube. Formation of violet to blue or green color (Liebermann–Burchard reaction for steroid) indicated the positive reaction. Formation of green color of the upper layer and reddish-brown color in the bottom layer confirmed a positive test for steroids and terpenoids respectively [17, 18].

Test for glycosides

The plant extract solution was dissolved in a mixture of glacial acetic acid (5%) and Ferric chloride solution and one or two drop of concentrated sulphuric acid was added. A brown ring or violet ring or greenish ring formation coloration indicated a positive test for glycosides [19].

One mL of distilled water was added to 10 drops of the extract dissolved in isopropyl alcohol (20 mg/mL) in a test-tube. The tube was shaken vigorously and observed for persistent froth formation and allowed to stand for 10 min. A positive result indicate a forth formation which is stable as long as 15 min [19].

Test for alkaloids

Few drops of Mayer's reagent (potassium mercuric iodide solution) was mixed with *I. mauritiana* extract solution. A yellowish-white like precipitation indicates a positive reaction for the presence of such metabolites [19].

Ferric chloride test for tannins

The plant extract solution of 10 mg was dissolved in ethanol (1 mL). Then distilled water (2 mL) was added followed by 4 drops of ferric chloride aqueous solution (10% w/v). The occurrence of phenols was confirmed by the development of dark blue or green color [20].

Total phenolic content

Total phenolic content of *I. mauritiana* extract was measured using a Folin-Ciocalteu colorimetric method spectrophotometrically as described by Idowu [21] with slight modification. Gallic acid was used as standard (concentration: 6.25–200 μ g/mL) while *I.*

mauritiana whole plant extract was 200 µg/mL. Firstly, 1 mL of the extract or standard gallic acid solution was used in screw cap tube and 5 mL of Folin-Ciocalteu reagent was added. Then, 4 mL (7.5%) of anhydrous sodium carbonate was added followed by 30 min incubation at 40 °C. The vehicle solvent was used as blank solution. UV absorbance was taken with a UV–VIS spectrophotometer (Shimadzu, Japan) at 765 nm. Total phenolic content was calculated as gallic acid equivalent (GAE) using the equation as follows:

$$C = (c \times V)/m$$

Where C = Total phenolics (mg/g plant extract in GAE), c = concentration of sample obtained from calibration curve (mg/mL), V = volume of the sample, and m = sample weight (g).

Total flavonoid content

To determine the total flavonoid content, the method of Rahman [22] was adopted, quercetin was the standard and different concentrations were used (6.25–200 µg/mL) while *I. mauritiana* methanol extract was 200 µg/mL. Initially, in a volume of 1 mL of extract or 1 mL quercetin solution of different concentrations, 3 mL of methanol was added. After that, 200 µL of 10% aluminum chloride and 200 µL of 1 M potassium acetate solution were added followed by the addition of 5.6 mL distilled water. Then the mixture incubated at room temperature for 30 min. Absorbance at 415 nm was recorded against the blank (water). The total flavonoid content was calculated as per the formulae below:

$$C = (c \times V)/m$$

Where C = Total flavonoid content (mg/g plant extract in Quercetin), c = concentration of sample obtained from calibration curve (mg/mL), V = volume of sample, and m = sample weight (g). Measurement was taken triplicate.

DPPH radical scavenging activity

To determine the antioxidant activity, the method of described by Braca [23] was adopted with slight modification. At first, 100 µL of plant extract and different concentrations of standard (ascorbic acid) were taken in test tubes. Then, 3 mL of 0.004% DPPH (2,2-diphenyl-1-picrylhydrazyl) solution in ethanol was added to each test tube followed by 30 min incubation. Later absorbance was taken at 517 nm spectrophotometrically against methanol (blank).

The absorbance values were used to calculate the percentage of antioxidant activity (% inhibition) using the formula below:

$$\% \text{Scavenging} = \{(A_0 - A_1)/A_0\} \times 100.$$

Where A_0 = Absorbance of DPPH solution only; A_1 = Absorbance in the presence of test sample and standard ascorbic acid in DPPH solution. Measurement was taken triplicate.

Antimicrobial activity of *I. mauritiana*

Microorganisms

Four gram-positive and six gram-negative bacteria were selected as test organisms. Gram positive; *Bacillus cereus*, *Bacillus subtilis*, *Sarcina lutea*, *Staphylococcus aureus* and Gram negative; *Escherichia coli*, *Salmonella typhi*, *Salmonella paratyphi*, *Pseudomonas aeruginosa*, *Vibrio parahemolyticus* and *Shigella dysenteriae* were used for antimicrobial activity test. Each of the stock cultures was collected as pure culture from the Institute of Nutrition and Food Science (INFS), University of Dhaka, Bangladesh.

Antibacterial activity

Disk diffusion method was used to perform the antibacterial test [24]. The sample solutions (400 and 200 µg/mL) were made by dissolving in methanol. Sterilized and dried filter paper discs (6 mm diameter) were then soaked in the crude extracts (400 and 200 µg/mL/disc) using micropipette and the solvent residue evaporated to dryness. Kanamycin (30 µg/disc) and blank discs (soaked with methanol followed by evaporation) were positive and negative control, respectively. The plates were inverted and refrigerated at 4 °C for 24 h for maximum diffusion. The following day plates were placed in the incubator at 37 °C for another 24 h for optimum bacterial growth. The antimicrobial activity was observed with inhibited growth of the microorganisms giving a clear, distinct zone of inhibition around the discs. Finally, the diameter of zone of inhibition was measured. The data was obtained from three individual experiments ($n = 3$).

Statistical analysis

A statistical analysis was used to interpret the antimicrobial and antioxidant results. The experiment was conducted in completely randomized design with 3 replicates. The results are presented as means ± standard error of means using MS excel [25].

Result and discussion

The results of preliminary phytochemical screening of *I. mauritiana* are presented in Table 1. Results revealed that

Table 1 Phytochemical screening of *I. mauritiana*

No.	Types of phytochemical constituents	Phytochemical composition (Methanolic extract)
1.	Terpenoids	+
2.	Saponins	+
3.	Flavonoids	+
4.	Glycosides	-
5.	Steroids	+
6.	Alkaloids	+

(+) = Present, (-) = Absent

the methanolic extract of *I. mauritiana* contains terpenoids, saponins, flavonoids, steroids, and alkaloids. The presence of these classes of compounds of this plant could be responsible for its various medicinal uses [26, 27].

On the other hand, the total phenolic content of *I. mauritiana* was evaluated as expressed by gallic acid equivalents per gram of extract which was found as 59.302 ± 3.289 mg of GAE/g. The value was obtained from regression equation of the calibration curve ($y = 0.004x + 0.105$; $r^2 = 0.992$) as shown in Fig. 1. Subsequently, the total flavonoid content of methanol extract of *I. mauritiana* was 27.212 ± 0.51 mg of QE/g which was obtained from the regression equation ($y = 0.003x + 0.023$; $r^2 = 0.995$) as shown in Fig. 2.

Similarly, DPPH free radical scavenging activity of *I. mauritiana* and ascorbic acid is shown in the Table 2. Both ascorbic acid and *I. mauritiana* methanolic extract showed dose dependent activity. Among the eight different concentrations (6.25, 12.5, 25, 50, 100, 200, 400 and 800 $\mu\text{g/mL}$) ascorbic acid showed 14.05 ± 1.64 , 16.66 ± 1.55 , 20.47 ± 1.47 , 27.20 ± 1.64 , 42.76 ± 1.64 , 74.89 ± 1.63 , 87.82 ± 1.73 and $90.96 \pm 1.64\%$ scavenging activity respectively while, *I. mauritiana* methanol extract showed 20.68 ± 1.15 , 26.50 ± 1.64 , 31.92 ± 1.80 , 38.45 ± 1.89 , 48.99 ± 1.73 , 59.03 ± 1.86 , 66.86 ± 1.78 and $72.28 \pm 1.8\%$ scavenging activity respectively. In addition, the IC_{50} of ascorbic acid and methanolic extract were

230.09 $\mu\text{g/mL}$ and 275.084 $\mu\text{g/mL}$ respectively as shown in Table 2. The DPPH scavenging activity of *I. mauritiana* methanol extract was lesser than that of standard ascorbic acid. The IC_{50} value denotes the minimum sample concentration required to scavenge 50% free radical. The antiradical activity of the extract may be due to their high content of phenols, as polyphenols which contribute to the antioxidant activity in living systems by chelation and electron transfer or hydrogen donating ability of hydroxyl groups in *ortho*- and *para*- positions thus neutralizing the free radicals [28, 29].

The results of antibacterial activity of the extract and standard kanamycin with respect to each of the test organisms are presented in Table 3. The mean zone of inhibition of kanamycin was between 37 to 42 mm and the extract which was between 13 to 19 mm, respectively at a concentration of 400 $\mu\text{g/disc}$. Zone of inhibition of standard was larger than extract. The extract showed the highest zone of inhibition against the gram positive; *Bacillus cereus* (19.25 ± 1.020 mm) and *Sarcina lutea* (18.5 ± 0.0813 mm) and gram negative *Shigella boydii* (18.25 ± 3.06 mm) and had no activity against *Escherichia coli*. Gram positive strains were more sensitive to the extract than the gram negative ones. However, the extract had the lowest antibacterial effect to *Pseudomonas aureus*. These results indicate that *I. mauritiana* extract is

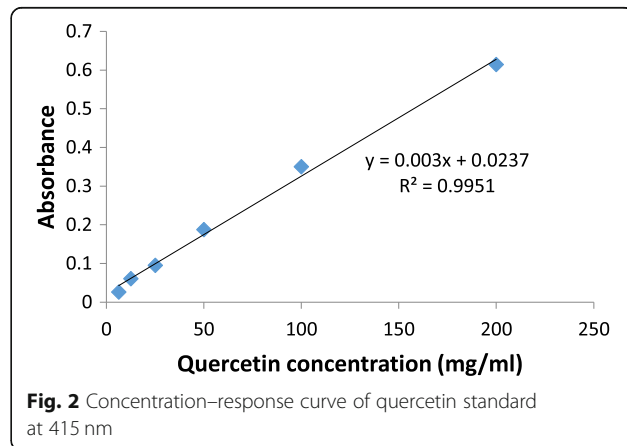
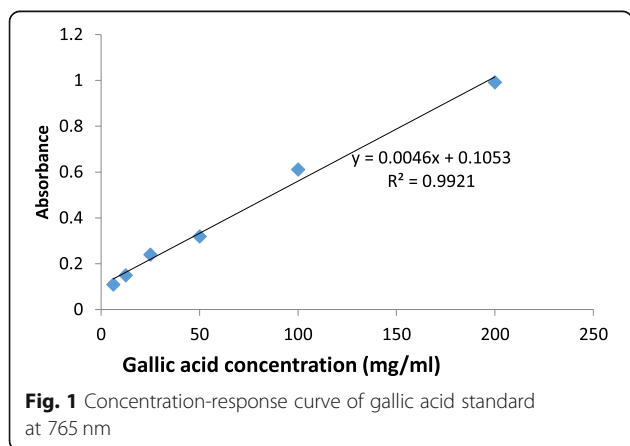


Table 2 DPPH free radical scavenging activity of ascorbic acid and *I. mauritiana* extract

Concentration (µg/mL)	% of scavenging of DPPH (Mean ± STD)	
	Ascorbic acid (Standard)	<i>I. mauritiana</i> extract
6.25	14.055 ± 1.637	20.682 ± 1.147
12.5	16.665 ± 1.551	26.506 ± 1.639
25	20.478 ± 1.471	31.927 ± 1.803
50	27.205 ± 1.636	38.453 ± 1.885
100	42.768 ± 1.635	48.996 ± 1.731
200	74.897 ± 1.636	59.036 ± 1.856
400	87.821 ± 1.726	66.867 ± 1.784
800	90.963 ± 1.638	72.289 ± 1.803
IC ₅₀ (µg/mL)	230.09	275.084

active against both bacterial strains as compared to standard antibiotic, kanamycin.

Plant has been a very significant source of medicine for decades and many plants have been tested for their potential bioactive compounds [30].

Free radicals are significant contributor in many pathological manifestations. Antioxidants can maintain good health and protect, either by counteracting these free radicals or defending the body's antioxidant defense mechanisms. Plants and herbs, being the potential sources for a number of antioxidants in addition to their therapeutic properties, have appealed the concern of scientific community to meet up the growing interest for raw materials with potential natural antioxidant compounds. Phenolic or polyphenolic compounds are one of the major classes of compounds having strong antioxidant properties. The antioxidative action of phenolic compounds is primarily due to their redox properties, [31, 32] which can hydrolyze and neutralize free radicals, purify singlet and triplet oxygen, or decompose peroxides.

On the other hand, ongoing demand for new antibiotic is present due to the sustained emergence of infection-resistant strains. About 80% of the drugs available in many developing countries originated from medicinal plants. In the industries of many countries plants comprise the raw material for processes that synthesizes pure chemical derivatives [33].

In the present study, prominent antioxidant activity was observed for the whole plant extract of *I. mauritiana* with potential antibacterial activity against a number of Gram (+) ve and Gram (-) ve bacteria. Different parts of the plant may vary with their antioxidant activity. For instance, a study revealed that, the root and stem bark, leaf and flower parts of *Tabebuia pallida* methanol extract were tested for antioxidant activity where the leaf extract was found to show highest antioxidative and free radical scavenging property which was also rich in total phenols and flavonoid contents [34]. The current study thus was conducted with the whole plant of *I. mauritiana*. It is clear from the results that, total phenolic and flavonoid contents and DPPH scavenging activity signifies the potential bioactivity of *I. mauritiana*. This result is consistent with the report of Sulaiman [35]. Moreover, phenolic compounds demonstrates potential antibacterial activity. Rahman and colleagues stated that, 3,4-dihydroxybenzoic acid obtained from *Cananga odorata* was active against a number of Gram (+) and Gram (-) bacteria [36].

In the same way, the antibacterial activity of *I. mauritiana* against gram positive bacteria such as *Bacillus cereus*, *Bacillus subtilis*, *Sarcina lutea*, *Staphylococcus aureus* and gram negative bacteria such as *Escherichia coli*, *Salmonella typhi*, *Salmonella paratyphi*, *Pseudomonas aeruginosa*, *Vibrio parahemolyticus* and *Shigella dysenteriae* was evaluated. These are important pathogens and can rapidly develop antibiotic resistance as

Table 3 Antibacterial activity of *I. mauritiana* using the disk diffusion method

Bacterial strains	Test organisms	Zone of inhibition (mm)		
		<i>I. mauritiana</i> (400 µg /disc)	<i>I. Mauritiana</i> (200 µg /disc)	Kanamycin (30 µg /disc)
Gram positive	<i>Bacillus cereus</i>	19.25 ± 1.020	–	42.56 ± 0.816
	<i>Bacillus subtilis</i>	14.25 ± 0.612	–	37.86 ± 1.224
	<i>Staphylococcus aureus</i>	–	–	41.25 ± 0.186
	<i>Sarcina lutea</i>	18.5 ± 0.0813	–	40.50 ± 0.408
Gram negative	<i>Escherichia coli</i>	–	–	39.66 ± 0.471
	<i>Pseudomonas aureus</i>	13 ± 0.816	–	40.05 ± 0.754
	<i>Salmonella paratyphi</i>	15.15 ± 0.122	–	41.48 ± 0.465
	<i>Shigella dysenteriae</i>	–	–	42.82 ± 0.275
	<i>Shigella boydii</i>	18.25 ± 3.06	–	41.95 ± 0.568
	<i>Vibrio parahemolyticus</i>	16 ± 0.816	–	38.52 ± 0.437

Note: The diameter of zone of inhibition are expressed as mean ± SD. (n = 3); a diameter less than 8 mm was considered inactive. Zone of inhibition (mm) determined after 24 h of incubation at 37 °C

antibiotic use increases. However, a previous report stated that, the leaf freeze dried extract of *I. mauritiana* did not show any activity against *Streptococcus mutans*, *S. mitis*, *Staphylococcus aureus*, and a fungi *Candida albicans* in both agar disk and agar well diffusion tests [37]. This study reveals the promise of antibacterial activity of *I. mauritiana* against gram positive bacteria in particular.

Conclusion

The results demonstrated that the methanolic extract of *I. mauritiana* Jacq shown antioxidant and antimicrobial activity. These findings could be a scientific evidence to use this plant as a potential source of antioxidant and antibacterial agents. However, future studies are necessary to determine the mechanisms of these pharmacologic properties. Moreover, phytochemicals characterization of possible bioactive compounds in *I. mauritiana* Jacq is also required.

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Authors' contributions

IA performed the extraction and antioxidant study and analyzed the data. MSF performed the antibacterial study. MSF and RM drafted the manuscript and MFFMA edited the manuscript. AKMMH planned and supervised the work and finalized the manuscript. All authors approved the final version of the manuscript.

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All data generated and analyzed are present in this manuscript.

Ethics approval and consent participate

None.

Consent for publication

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Competing interests

Authors have no conflict of interest.

Author details

¹School of Science and Engineering, Department of Pharmacy, Southeast University, Dhaka, Bangladesh. ²Faculty of Industrial Sciences & Technology, Universiti Malaysia Pahang, Lebuhraya Tun Razak, 26300, Gambang, Pahang, Malaysia. ³School of Medicine, Department of Pharmacy, University of Asia Pacific, 74/A, Green Road, Dhaka, Bangladesh.

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