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Phytochemical profiling, antioxidant activity, antimicrobial activity and GC-MS analysis of *Ipomoea aquatica* Forsk collected from EMA market, Manipur

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Abstract
The present study was carried out to analyze the phytochemical constituents, antioxidant activity, total phenolic content, total flavonoid content and antimicrobial activity of Ipomoea *aquatica* Forsk. Preliminary phytochemical analysis was done using standard procedure. Total phenolic content, total flavonoid content and antioxidant activities were determined spectrophotometric ally in crude aqueous and methanolic extracts. The *in vitro* antimicrobial activity was determined by well diffusion method against four MTCC bacterial species and three MTCC fungal species. Preliminary phytochemical analysis indicated the presence of most of the phytochemicals except steroid and phlobatannins. Antioxidant activities increased with increase in concentration of the extract. The methanolic extract showed a better result than the aqueous extract. Clear antimicrobial activity was observed against *Staphylococcus aureus* and *Escherichia coli* using methanolic extract while aqueous extract showed moderate activity. Moderate activity was noted against *Pseudomonas aeruginosa* using methanolic extract whereas the aqueous extract was found to be inactive towards the same organism. Antifungal activity species was absent. GC-MS analysis was carried out to identify the bioactive compounds in methanolic extract as methanolic extract was showing a better result. The present study demonstrates the antioxidant and antimicrobial activity of *Ipomoea aquatica* Forsk due to the various phytochemicals present in the extract which confer their traditional uses as food and medicine.

Keywords: *Ipomoea aquatica* Forsk, phenolic, flavonoids, antioxidant, antimicrobial and GC-MS

Introduction
Ipomoea aquatica Forsk (*I. aquatica* Forsk) belonging to Convolvulaceae family is a commonly grown green leafy vegetable found throughout India, Ceylon, Tropical Asia, Africa and Australia. This plant is grown as a semi-aquatic plant and found abundantly in marshy areas. It is locally known as Kolamani in Meiteilon (Manipuri). *I. aquatica* Forsk is a rich source of vitamins, minerals, proteins, fibers, carotenes and flavonoids with many health benefits (Manvar and Desai, 2013) [1]. In Ayurveda, it is recommended to consume *I. aquatica* Forsk to mitigate disorders like jaundice. The effectiveness of these plant products from traditional claims must be proved to help develop novel drugs acting against these disorders. Traditionally, *I. aquatica* Forsk used as a carminative agent. It has also been found to lessen inflammation, and is useful in fever, jaundice, biliousness, bronchitis, liver complaints, etc. Various parts of the *I. aquatica* Forsk plant are used medicinally in Southeastern Asia and reported to be useful for the treatment of high blood pressure, as an emetic in the treatment of opium and arsenic poisoning (Perry, 1980) [2].The dried juice has been reported to be a purgative, while the leaves and stems were found to possess cooling action. Moreover, it is also traditionally used in the treatment of nervous and general debility, piles, worm infections, leucoderma, leprosy, jaundice and liver complaints (Chopra *et al*., 1956; Vickers and Zollman, 1999) [3, 4].

Very few studies have been done in this plant. This includes the inhibition of prostaglandin synthesis, eye diseases, constipation and hypoglycemic effects (Tee and Lim, 1991) [5]. Phytochemical investigations of this plant have revealed the presence of carotenes such as β-carotene, cryptoxanthin, lutein, lutein epoxide, violoxanthin and neoxanthin, flavonoids such as mycertin, quercetin, luteolin and apigenin and some alkaloids (Tofern, 1999) [6].

In Manipur, *I. aquatica* Forsk is commonly consumed as vegetable in fresh and cooked form. It is available almost throughout the year mainly during the rainy seasons. Noticing the frequent consumption of this plant with its reported medicinal properties, the present study

Was carried out to analyze the phytochemical components, antioxidant and antimicrobial activities of the crude aqueous and methanolic extracts of *I. aquatica* Forsk. GC-MS analysis of the methanolic extract was further carried out to identify the presence of bioactive components.

Materials and Methods

Plant sample

Ipomoea aquatica Forsk was brought from Ema market of Manipur, Northeast India. Identification of the sample was done by L. Somarjit Singh, Associate Professor, Department of Botany, Imphal College, Imphal. Stem and leaves of the plant were washed with tap water and then rinsed with distilled water, shade dried and ground into fine powder.

Soxhlet extraction

40g of powdered *I. aquatica* Forsk was extracted separately using 400ml of methanol and double distilled water by soxhlation until the solvent become colourless in main chamber of the soxhlet extractor. The extracts were evaporated to dryness and crude extracts were obtained. The crude extracts were screened for the phytochemical constituents.

Phytochemical screening was carried out for crude aqueous and methanolic extracts of *I. aquatica* Forsk using standard protocol (Audu *et al*., 2007; Edeoga *et al*., 2005; Kokate, 2005; Tiwari *et al*., 2011; Rathod and Valvi, 2011; Bag *et al*., 2016; Bhaigyabati *et al*., 2017) [7,8,9,10,11,12,13].

Determination of total phenolic content

The amount of total phenolic content in crude aqueous and methanolic extracts of *I. aquatica* Forsk was determined with Folin-Ciocalteu method (Spanos and Wrolstad, 1990; Lincoln, 2001; Chakraborthy and Ghorpade, 2010; Bag *et al*., 2016; Bhaigyabati *et al*., 2017) [14, 15, 16, 12, 13].

Estimation of total flavonoid content

Total flavonoid content in the sample extracts were estimated by aluminium chloride colorimetric method (Akbay, 2003; Kaufman *et al*., 1999; Bag *et al*., 2016; Bhaigyabati *et al*., 2017) [17, 18, 12, 13].

Determination of free radical scavenging assay

The free radical scavenging capacities of the crude aqueous and methanolic extracts of *I. aquatica* Forsk were determined using DPPH method (Braca *et al*., 2001; Bag *et al*., 2016; Bhaigyabati *et al*., 2017) [19, 12, 13].

Estimation of reducing power

Reducing power of various concentrations (20-100µg/ml) of the crude aqueous and methanolic *I. aquatica* Forsk extracts were determined by Ferric reducing anti-oxidant power assay. Increased absorbance of the reaction mixture indicates increase in reducing power (Oyaizu, 1986; Bag *et al*., 2016; Bhaigyabati *et al*., 2017) [20, 12, 13].

Determination of total antioxidant activity

The phosphomolybdenum method was used to evaluate the total antioxidant activity of the extracts (Prieto *et al*., 1999; Bag *et al*., 2016; Bhaigyabati *et al*., 2017) [21, 12, 13].

Antimicrobial assay

Test organisms

The test bacteria used were the Gram positive organisms *Staphylococcus aureus* (MTCC 737), and *Bacillus subtilis* (MTCC 441), and the Gram negative bacteria *Escherichia coli* (MTCC 738) and *Pseudomonas species* (MTCC 424). The test fungi used were *Fusarium oxysporum* (MTCC 227), *Trichoderma viride* (MTCC 793) and *Aspergillus Niger* (MTCC 281). All the reference strains were procured from Microbial Type Culture Collection (MTCC), Institute of Microbial Technology (IMTECH), and Chandigarh, India.

Media

Nutrient Agar (NA) and Nutrient Broth (NB) were used for bacterial culture and Potato Dextrose Agar (PDA) and Potato Dextrose Broth (PDB) were used for fungal culture in each experiment.

Antimicrobial agents

Streptomycin (10 µg) and Kanamycin (30 µg) discs were used as positive control for bacteria and fungi respectively and sterile distilled water was used as negative control.

Preparation of extract of different concentration

Stock solution of each of the crude extracts was prepared with N, N-Dimethyl sulfoxide (DMSO) making a concentration of 40 mg/ml. From the stock, three different dilutions were made to obtained 2 mg/50 µl 1 mg/50µl and 0.5 mg/50µl for both the aqueous and methanolic extracts.

Antimicrobial activity

Antimicrobial activity was done by using Kirby-Bauer method (Bauer *et al*., 1966) [22]. A loopful of freshly grown test organisms (both bacteria and fungi) were inoculated on NB (for bacteria) and PDB (for fungal) in an orbital shaker (150 rpm, 30°C). The test bacteria were incubated for 24 hrs while the test fungi were incubated for 48-72 hrs. 0.1 ml each of the bacterial and fungal broths was spread uniformly with a sterile L shaped spreader on NA and PDA plates respectively. Wells were punched with the help of a cork borer (6 mm diameter) on the media plates. Then 50 µl each of 500 µg, 750 µg and 100 µg concentrations were put in each well in triplicates for both the extracts and incubated for 24-48 hrs at 30°C. The zone of inhibition (in mm diameter) were measured (as mean of the triplicate readings) using a scale [22] and taken as the activity against the test organisms. The zone of inhibition was graded according to Kang SN *et al*., 2013 [23] (Table 1). Streptomycin (10 µg) and Kanamycin (30 µg) discs were used as positive control for bacteria and fungi respectively and sterile distilled water as negative control. These plates were kept at 4 °C for 1 hour for proper diffusion of the test organisms.

Table 1: Grading of zone of inhibition

Diameter of ZOI	Antimicrobial activity
6 - 8 mm	No antimicrobial activity
8.1 - 9 mm	Slight antimicrobial activity
9.1-12 mm	Moderate antimicrobial activity
12.1 – 15 mm	Clear antimicrobial activity
> 15 mm	Strong antimicrobial activity

GC-MS Analysis

Gas chromatography (GC) analysis was carried out at Advanced Instrumentation Research Facility (JNU) New Delhi. This technique is best for identification of various phytochemicals of plant extract. The equipment used was GC-MS QP-2010 ultra. The carrier gas used in GC-MS programme was helium at 1 ml/minute (split ratio=10:0). The initial oven temperature program is 50°C and final

temperature is 280 ℃ withhold time of 22 min, Ion Source temperature is 220 ℃ and interface temperature is 270 ℃, solvent cut time 5.50 min, Detector Gain Mode: Relative to the tuning result, Detector Gain +0.00 KV, threshold 1000, start time 6.0 min, end time 44.98 min, Event time 0.50 sec, Scan speed 1250, start m/z 50.00 and end m/z 650.00.

Result and Discussions

The preliminary phytochemical screening of the crude aqueous and methanolic extracts of *I. aquatica* Forsk is indicated in Table 2. Preliminary phytochemical screening shows the presence of most of the phytochemicals in both aqueous and methanolic extracts of *I. aquatica* Forsk except steroids and phlobatannins. Presence of cardiac glycosides was noted in aqueous extract while the presence of terpenoids and oil was observed in methanolic extract.

Table 2: Phytochemicals present in crude aqueous and methanolic extracts of *I. aquatica* Forsk

Phytochemical constituents	Test	*Ipomoea aquatica* Forsk edible part	
		Aqueous extract	Methanolic extract
Amino acids	Ninhydrin test	-	-
Alkaloids	Hager's test	+	+
Carbohydrates (reducing sugar)	Benedict's test Fehling's test	+ +	+ +
Proteins	Xanthoproteic test	+	+
Flavonoids	Alkaline reagent test Lead acetate test	+	+
Phenolic compounds	Lead acetate test Ferric Chloride test	+	+
Steroids and Terpenoids	Salkowski's test	- -	- +
Saponins	Froth test	+	+
Tannins	Lead acetate test Ferric chloride test	+ +	+ +
Cardiac glycosides	Keller-killiani test	+	-
Oil		-	+
Phlobatannins		-	-

Key: + = presence and - = absence

Similar result was reported by Das *et al*., 2018 [24] indicating the presence of flavonoids, saponins, tannins and steroids in the aqueous and methanolic extracts of *I. aquatica* Forsk. While Shamli and Chandra, 2015 [25] have showed the presence of glycosides, flavonoids, phenols, tannins and terpenoids in acetone extract of *I. aquatica* Forsk.

Our result showed methanolic to be a better solvent for the extraction of phytochemicals from *I. aquatica* Forsk and also found that *I. aquatica* Forsk is a rich source of phytochemicals. Preliminary phytochemical screening is usually performed for the identification of generous phytochemicals which may be responsible for the antioxidant and antimicrobial activity of plant extracts (Sagbo *et al*., 2017) [26].

Total phenolic content was estimated in crude aqueous and methanolic extracts of *I. aquatica* Forsk by Folin-Ciocalteu method. Standard curve of Gallic acid is illustrated in fig. 1. Total phenolic content in the sample extracts were calculated using the above mentioned formula and data obtained from the Gallic acid calibration curve (y= 0.010x, R^2= 0.999). Total phenolic content in crude aqueous and methanolic extracts were found to be 4.014 ± 0.002 mg/g (GAE) and 9.742 ± 0.03mg/g (GAE) respectively indicating a higher total phenolic content in methanol extract than aqueous extract.

Total flavonoid content in crude aqueous and methanolic extracts of the plant sample was quantified by Aluminum chloride method. The Quercetin standard calibration curve is shown in fig. 2.

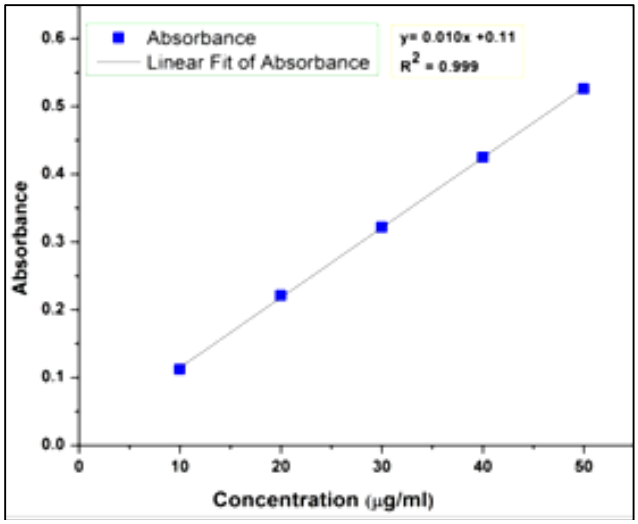


Fig 1: Standard curve of Gallic acid

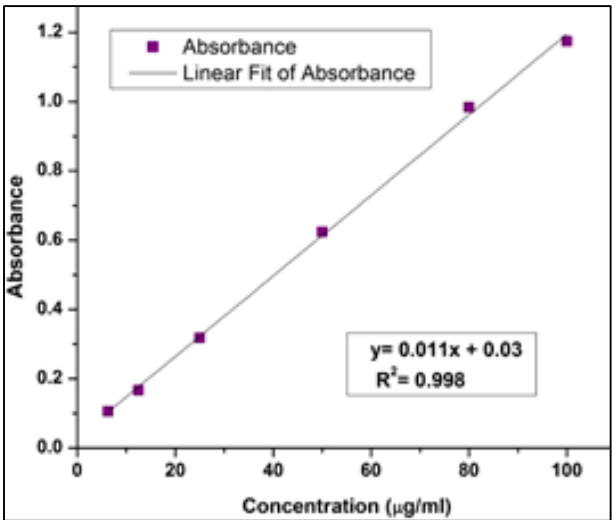


Fig. 2: Standard curve of Quercetin

From the standard curve of quercetin (y= 0.011x, R^2= 0.998), concentration values of both extracts were obtained and total flavonoid content (TFC) was calculated by using the following formula (Chang *et al*., 2002) [27].

TFC = R * D.F * V * 100 / W

Where R - Result obtained from the standard curve, D.F - Dilution factor, V - Volume of stock solution, 100 - For 100 g dried sample and W - Weight of plant sample used in the experiment.

Total flavonoid content was found to be higher in methanolic extract (11.26 ±0.02 µg/100g) than aqueous extract (7.95 ± 0.02µg/100g) of the plant. Table 3 indicates the total phenolic and total flavonoid content in crude aqueous and methanolic extracts of *I. aquatica* Forsk.

Table 3: Total phenolic and total flavonoid content in crude extracts of *I. aquatica* Forsk

Sample	Total phenolic content in mg /g of extract (in GAE)		Total flavonoid content in µg/100g of dried extract (in QE)	
	Aqueous	Methanol	Aqueous	Methanol
Ipomoea aquatica Forsk edible part	4.014 ± 0.002	9.742 ± 0.03	7.95 ± 0.02	11.26 ±0.02

Assays were performed in triplicates. Values are expressed as means ± SD

Our result showed methanol a better solvent for extraction of phenolic compounds from *I. aquatica* Forsk when compared to water. Methanol has been generally found to be more efficient in extraction of phytochemicals.

Thi and Hwang, 2015 [28] found that 80% ethanol extract of *I. aquatica* Forsk contains 35.6mg/g total polyphenols and 40.4 mg/g total flavonoid. Difference in the polyphenolic content from our result may due to the difference in geographical condition and also to the solvent used in extraction of phytochemicals.

Phenolic compounds are the most abundant phytochemical in plants and considered as important natural antioxidants. Phenolic compounds exhibit their antioxidant activity by various mechanisms such as donation of hydrogen atoms to free radicals and through connection to transition metal ions resulting in more stable forms (Kumar *et al*., 2014) [29]. Various physiological actions performed by polyphenols were related to the prevention of neurodegenerative and cardiovascular diseases, cancer, among others, mainly because of their high antioxidant capacity (Wootton-Beard *et al*., 2011) [30].

DPPH is a free radical compound and has been widely used to test the free radical-scavenging ability of various samples (Sakanaka *et al*., 2005) [31]. It is a stable free radical with a characteristic absorption at 517 nm that was used to study the radical-scavenging effects of extracts. As antioxidants donate protons to this radical, the absorption decreases. Antioxidants, on interaction with DPPH, transfer either an electron or hydrogen atom to DPPH, thus neutralizing its free radical character (Naik *et al*., 2003) [32]. The color changed from purple to yellow and the absorbance at wavelength 517 nm decreased.

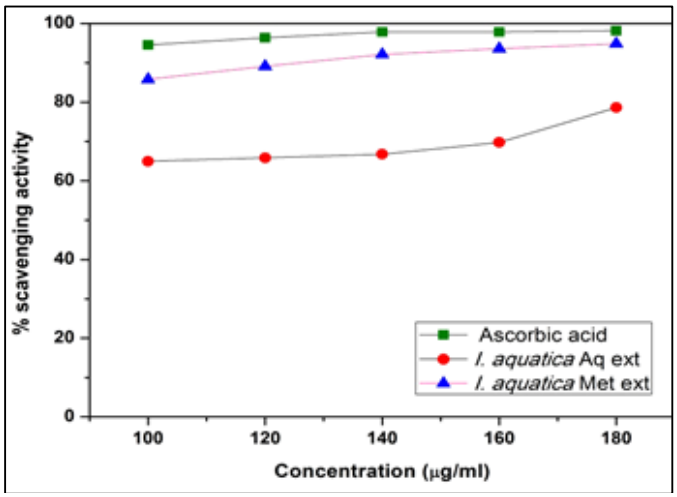


Fig 3: DPPH scavenging activity shown by standard and crude extracts of *I. aquatica* Forsk

DPPH assay showed an increase in concentration increases the free radical scavenging activity for the reference standard, ascorbic acid and crude aqueous and methanolic extracts of *I. aquatica* Forsk. Percentage DPPH scavenging activity of aqueous and methanolic extracts was comparable with standard ascorbic acid and methanolic extract showed higher scavenging activity than aqueous extract.

Reducing power assay indicates an increasing order for standard as well as for both the extracts of plant sample and is shown in fig. 4.

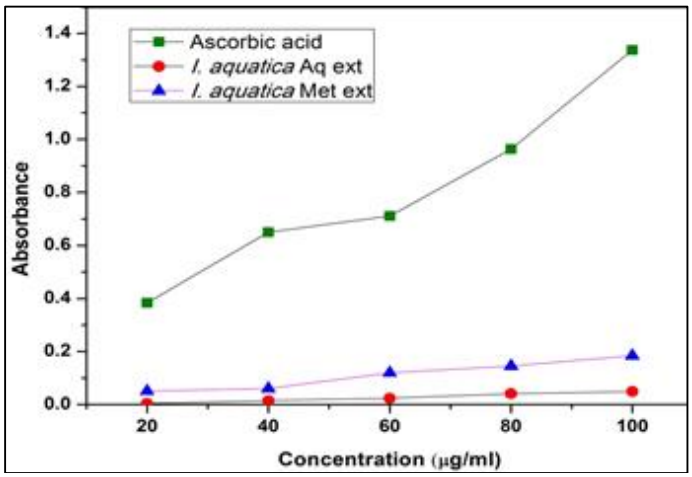


Fig 4: Reducing power shown by standard and crude extracts of *I. aquatica* Forsk

Reducing power noted in aqueous and methanolic extract is almost equal with methanolic extract showing a slight higher in reducing power than aqueous extract of the plant. Reducing power of the extracts may be contributed by bioactive compounds in the extract which possess electron donating abilities. Presence of reducers causes the conversion of the Fe^{3+} complex to the ferrous (Fe^{2+}) form which serves as a significant indicator of its antioxidant capacity (Yildirim *et al*., 2000) [33].

Similar trend was noted in total antioxidant activity where methanolic extract showed higher activity than aqueous extract. Total antioxidant activity shown by various concentrations (20-100 µg/ml) of crude aqueous and methanolic extracts of *I. aquatica* Forsk is shown in table 4.

Table 4: Total antioxidant activity of *I. aquatica* Forsk

Concentration (µg/ml)	Total antioxidant activity in µg/ml of extract (in AAE)	
	Ipomoea aquatica Forsk edible part	
	Aqueous	Methanol
20	2.4± 0.02	10.2± 0.01
40	4.8± 0.05	11.4± 0.02
60	5.4± 0.02	17.4± 0.03
80	7.5± 0.01	21.3± 0.04
100	8.1± 0.03	27.9± 0.02

Assays were performed in triplicates. Values are expressed as means ± SD

At highest concentration used for the study, total antioxidant activity shown by methanolic extract was 27.9µg AAE/mg of extract while that of aqueous extract was 8.1µg AAE /mg of extract.

Prasad *et al*., 2005 [34] have isolated a free radical-scavenging antioxidant from methanol extract of *I. aquatica* Forsk which has exhibited a strong antioxidant activity. Thi and Hwang, 2015 [28] has reported that DPPH scavenging activity and reducing power assay of *I. aquatica* extract were increased in a concentration dependent manner which is similar to the result we found. As per Hwang *et al*., 2011 [35] phenolic and flavonoid content was higher in ethanol extract than aqueous extract and also showed higher DPPH scavenging activity and reducing power.

Antimicrobial Screening of both the methanolic and aqueous extracts of *I. aquatica* Forsk showed that both the extracts (Fig 5 and Fig 6) were found to have antibacterial activity in one or the other extracts against *E. coli* (MTCC 738), *P.aeruginosa* (MTCC 424) and *S. aureus* (MTCC 737) but no activity was observed against *B. subtilis* (MTCC 441). Both the extracts exhibit no antifungal activity against any of the test fungi. The methanolic extract exhibited more antibacterial activity when compared with the aqueous extract. No antimicrobial activity was observed against *P.aeruginosa* (MTCC 424) for all the different concentrations in aqueous extract but moderate activity was observed in the methanolic extract at 750µg/50µl and 1000 µg/50µl. Slight antibacterial activity was found against *S. aureus* (MTCC 737) for the methanolic extract at 500µg/50µl and becomes moderate at 750µg/50µl but the activity becomes clear when the concentration increases to 1000 µg/50µl. At the highest concentration for aqueous extract slight antibacterial activity was observed against *S. aureus* (MTCC 737) though at lower concentration no activity was observed.

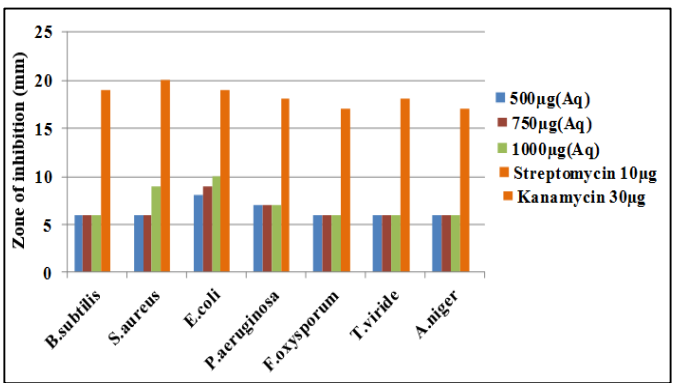


Fig 5. Antimicrobial activity of aqueous extract of *I. aquatica* Forsk against the test organisms

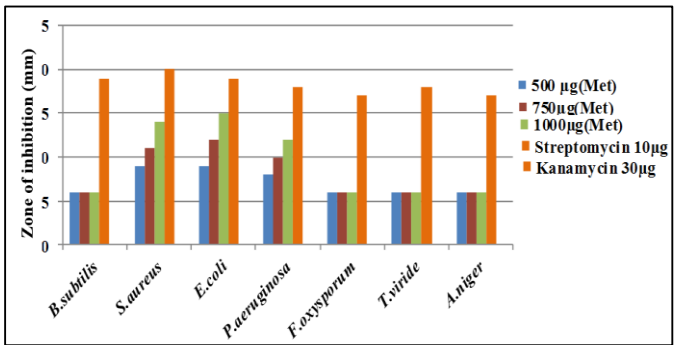


Fig 6. Antimicrobial activity of methanolic extract of *I. aquatica* Forsk against the test organisms

The activity against *E. coli* (MTCC 738) was found to be potent at the highest concentration for methanolic extract, though the activity was moderate for aqueous extract. Thus, it was observed that the antibacterial activity increased with the increase in concentrations of the extracts. Aqueous extract of *I. aquatica* Forsk showed lesser or no antibacterial activity when compared with methanolic extract which showed moderate to strong activity. Different solvents have been reported to have the capacity to extract different phytoconstituents depending on their solubility or polarity of the solubility or polarity in the solvent. Methanolic extracts have been reported to have higher solubility for more phytoconstituents, consequently showed higher antibacterial activity (Cowan, 1999) [36].

GC-MS is the best technique to identify the bioactive constituents of long chain hydrocarbons, alcohols, acids, esters, alkaloids, steroids, amino acid nitro compounds, etc. GC-MS analysis of the methanolic extract of *I. aquatica* Forsk reveals the presence of number of compounds from the GC fractions and these compounds were identified with mass spectrometry attached to GC.

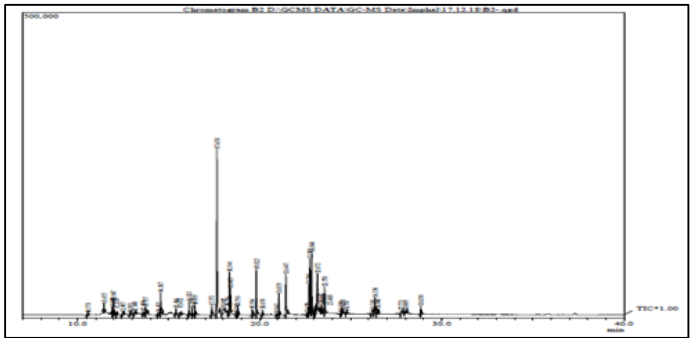
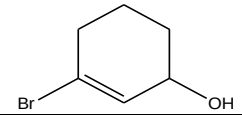
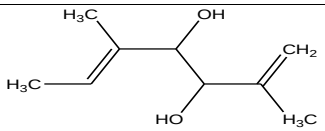
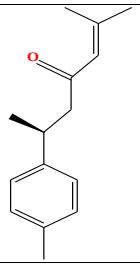
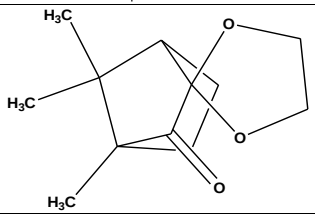
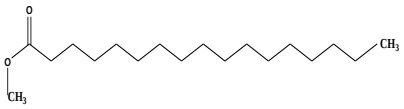
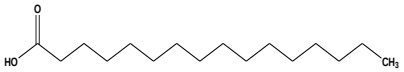
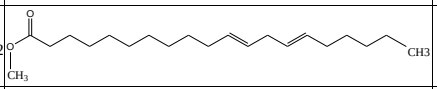
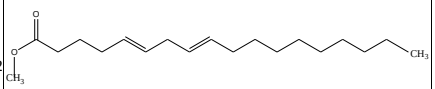
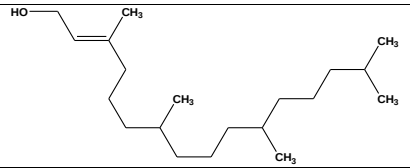
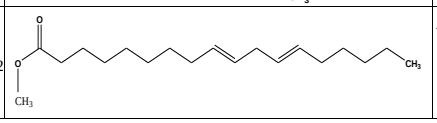
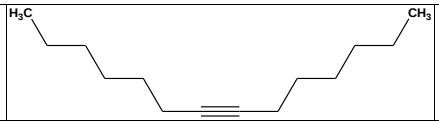


Fig 7: GC-MS chromatogram of methanolic extract of *I aquatica* Forsk

In the present study, 40 compounds have been identified from methanolic extract of the edible parts of *I. aquatica* Forsk by Gas Chromatography-Mass Spectrometry (GC-MS). Among the 40 compounds, 11 compounds were found to be major compounds, viz: 1,5-Heptadiene-3,4-diol, 2, 5- dimethyl, 9,12-Octadecadienoic acid, methyl ester, (E,E), phytol, hexadecanoic acid, 5,8-Octadecadienoic acid, methyl ester, Ar-tumerone, 7-Tetradecyne, 2-Cyclohexen-1-ol, 3-bromo, 11,14-Eicosadienoic acid, methyl ester and Heptadecanoic acid, methyl ester. The identified compounds and their retention time, peak area (%), molecular weight, molecular formula, structure and activities related to medicinal uses are given in table 5.

Table 5: Major compounds identified in the methanolic extract of *I aquatica* Forsk

Sl.no.	Name of the compounds	RT	Area %	M.W	M.F	Structure	Medicinal uses
1.	2-Cyclohexen-1-ol, 3-bromo-	14.567	3.57	177.041	C_6H_9BrO		No activity reported
2.	1,5-Heptadiene-3,4-diol,2,5-dimethyl	17.658	21.92	156.225	$C_9H_{16}O_2$		No activity reported
3.	Ar-tumerone	17.373	4.18	216.324	$C_{15}H_{20}O$		Antidermatophytic (Alqasoumi *et al.*, 2012) [37]
4.	Spiro [bicyclo [2.2.1]heptane-2,2'- [1,3] dioxolan]-3-one, 4,7,7-trim ethyl-	19.825	5.51	210.273	$C_{12}H_{18}O_3$		No activity reported
5.	Heptadecanoic acid, methyl ester	21.059	2.24	284.484	$C_{18}H_{36}O_2$		Antibacterial and antifungal (Khasawneh *et al.*, 2011) [38], Anti-inflammatory, Hypercholesterolemic, cancer preventive, hepatoprotective, nematicide, insectifuge, antihistamine, antieczemic, antiacne, alpha reductase inhibitor, antiandrogenic, antiarthritic, anticoronary (Dagla *et al.*, 2012) [39].
6.	Hexadecanoic acid	21.447	6.30	270.45	$C_{17}H_{34}O_2$		Anti-inflammatory (Jankasem *et al.*, 2013) [40], Antioxidant, Hypercholesterolemic nematicide, pesticide, anti-androgenic flavor, hemolytic, 5-Alpha reductase inhibitor (Chandrasekaran *et al.*, 2011) [41] potent mosquito larvicide (Krishnamoorthy and Subramaniam, 2014) [42]
7.	11,14-Eicosadienoic acid, methyl ester	22.700	2.58	322.533	$C_{21}H_{38}O_2$		No activity reported
8.	5,8-Octadecadienoic acid, methyl ester	22.762	5.99	280.452	$C_{18}H_{32}O_2$		Anti-inflammatory, hypocholesterolemic, cancer preventive, hepatoprotective, nematicide, insectifuge, antieczemic, anticancer, antiarthritic insectifuge, antihistaminic, anticoronary (Khasawneh *et al.*, 2011) [38].
9.	Phytol	22.861	6.96	296.539	$C_{20}H_{40}O$		Anticancer, antioxidant, anti-inflammatory, diuretic, antitumor, chemo preventive, antimicrobial, use in vaccine formulations (Khasawneh *et al.*, 2011) [38].
10.	9,12-Octadecadienoic acid, methyl ester, (E,E)-	23.172	7.00	294.4721	$C_{19}H_{34}O_2$		Anti-cancer and anti-inflammatory (Krishnamoorthy and Subramaniam, 2014 ;Khasawneh *et al.*, 2011) [42, 38]

11.	7-Tetradecyne	23.570	4.00	196.378	$C_{14}H_{28}$		No activity reported

The antioxidant activity shown by *I. aquatica* Forsk may be due to presence of two compounds, namely, hexadecanoic acid and phytol. The antimicrobial activity of *I. aquatica* Forsk may be due to presence of phytol which is known for its antimicrobial activity. The phytol is the compound which inactivates the protein and enzymes present in the microorganisms. In addition, the compounds have no remarkable toxicity and possess high stability (Aparna *et al*., 2012; Kumar *et al*., 2010; Rahuman *et al*., 2000) [43, 44, 45]. Thus, from the present finding it is evident that phytol along with other phytoconstituents may be responsible for the inhibition of the growth of *S. aureus* and *E. coli* in methanolic of *I. aquatica* Forsk.

Conclusion

The present study showed methanolic extract of *Ipomoea aquatica* Forsk has higher antioxidant and antimicrobial activities than aqueous extract which may be due to higher total phenolic content and total flavonoid content in the methanolic extract, which would justify its traditional use. GC-MS result also reveals the presence of 11 major compounds in methanolic extract, which may be responsible for the antioxidant and antimicrobial activities of the extract. Further studies can be done to find out the biological activity of major compounds whose activities have not been reported so far.

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