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PHYTOCHEMICAL AND ANTIMICROBIAL EVALUATION OF *MALACHRA* *CAPitata* (L) L EXTRACTS

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ABSTRACT

Preliminary phytochemical screening in various solvent extracts of *Malachra capitata* root, stem and leaf dry powder samples revealed the presence of alkaloids, anthocyanoides, coumarins, flavonoids, phenol, protein, tannin and terpenoids. The quantitative analysis of various biomolecules in the different parts of *Malachra capitata* shows maximum amount of reducing sugar, protein and lipid in leaf, while maximum carbohydrate and starch content was noted in stem of *Malachra capitata* as compared to other samples. Quantitative analysis of antioxidant (total phenol, total tannin and vitamin- C) in different parts of *Malachra capitata* shows maximum amount of these compounds in leaf sample when compared to other two samples of root and stem. Antimicrobial activity of *Malachra capitata* root, stem and leaf samples in ethanol extract shows potential activity against the growth of the bacterial (*Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus subtilis*,) and fungal strain (*Candida albicans*, *Candida parapsolisis* and *Aspergillus niger*) tested.

1. INTRODUCTION

Phytochemical evaluation is used for quality assessment of plants which includes preliminary phytochemical screening. In order to discover new bioactive compounds, the plant extracts are evaluated by chemical screening¹. Plant derived natural products such as flavonoids, terpenes and alkaloids^{2, 3} triterpine derivatives, anthroquinon derivatives, polyphenolics, comprising flavonoids, colchicines and proanthocyanidins⁴ and compounds like peptides, unsaturated ion-chain fatty acids, aldehydes, essential oils, phenols and water or ethanol soluble compounds are also significant in therapeutic application against human and animal pathogen, including bacteria, fungi and viruses^{5, 6}. Members of Malvaceae family are widespread⁷ and are known by their different uses in folk medicine such as diuretic, in treatment of rheumatism, gastrointestinal disorders^{8, 9}, anti-inflammatory and antinociceptive¹⁰, snakebites¹¹ and asthma¹². The active components of the Malvaceae species are found in the leaves, flowers, seeds and roots. The root of the *M. capitata* is a traditional remedy for the many disease condition such as pain, hepatic cirrhosis, inflammation, diarrhea, convulsion, dementia, pyrexia, ulcer and healing of wounds¹³⁻¹⁵. In Punjab, *M. capitata* plants are traditionally used for curing blood infection in cattles, dental problem, joint pain and wounds¹⁶. Previous phytochemical investigations of Malvaceae species describe the isolation of alkaloids, anthocyanins, flavonoids, phenolic acids, saponins, steroids, glycosides, tannins, triterpenes and volatile oils^{9, 17-22}. The present study was carried out to evaluate phytochemical constituents and the antimicrobial activity of *Malachra capitata* root, stem and leaf sample extracts.

2. MATERIALS AND METHODS

2.1. Source of Plant Material. *Malachra capitata* was collected from Komaneri village of Thoothukudi district, Tamil Nadu, India and identified by Dr. Chelladurai, Research Officer, at Central Council for Research in Ayurveda and Siddha, Palaymkottai.

2.2. Preparation of dry powder samples of *Malachra capitata*: The different parts (root, stem and leaf) of *M. capitata* collected were dried separately at room temperature ($30^{\circ}\text{C} \pm 2^{\circ}\text{C}$) for about two weeks to get a constant weight. The dried plant materials were ground to powder by mechanical device and subjected to qualitative and quantitative phytochemical analysis.

2.3. Preliminary qualitative phytochemical screening: The different parts of *M. capitata* dry powder samples were extracted with different solvents such as petroleum ether, chloroform, ethyl acetate, methanol, ethanol and water at 20% (w/v) level in a soxhlet apparatus. The extracts were concentrated and used for qualitative phytochemical analysis of alkaloids,

anthocyanoids, coumarins, flavonoids, phenols, proteins, quinine, saponin, steroids, sucrose, tannins, and terpenoids following the standard methods^{23, 24}. Phytocompounds such as carbohydrates²⁵, reducing sugar²⁶, starch²⁷, Protein²⁸, total lipid²⁹⁻³¹, antioxidants such as total phenol, tannin³² and vitamin-C³³ were quantitatively analyzed following standard methods.

2.4. Antimicrobial assay

2.4.1. Preparation of plant extracts for antimicrobial activity: Ethanol was used for the extraction of the active components present in the different parts of *M. capitata* root, stem and leaf. For ethanol extraction, 60g of the air dried powder was extracted with ethanol (40-60°C) in a Soxhlet extractor for 18-20h and solution was evaporated to dryness under reduced pressure and controlled temperature by using rotoevaporator. The extract was stored in a refrigerator at 4°C in air tight bottles until further use.

2.4.2. Microorganisms used: Three bacterial and three fungal were used in this study. Standard strain of *Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus subtilis* (bacterial), *Candida albicans*, *Candida parapsolis* and *Aspergillus niger* (fungal) organisms were used. The antimicrobial agents Netilmycin (30mcg) and Flucanazole (25mcg) were used as positive control for the present study.

Nutrient agar medium was prepared and autoclaved at standard temperature of 121°C, pressure of 15psi (pounds per square inch) for a time period of 15 minutes. The autoclaved medium was aseptically transferred into pre sterilized Petri plates which are allowed to cool for solidification of medium. Now in aseptic conditions, on the solid agar surface, culture of *Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus subtilis*, *Candida albicans*, *Candida parapsolis* and *Aspergillus niger* inoculated by spread plate method in different Petri plates respectively. Thereafter, under aseptic conditions, saturated filter paper discs were placed on the inoculated solid agar surface. Similarly, commercially available Netilmycin (30mcg) and Flucanazole (25mcg) antibiotic discs were placed aseptically on the inoculated solid agar surface as a positive control for bacteria and fungi respectively. The Petri plates were finally marked accordingly and incubated at 37°C for 48 hours. Antimicrobial activities of the extracts were determined by measuring the diameter of inhibitions zone (mm) against each microbe tested.

3. RESULTS AND DISCUSSION

3.1. Qualitative preliminary phytochemical analysis: Plants produce primary and secondary metabolites which encompass a wide array of functions³⁴. Primary metabolites are necessary for cellular processes, while secondary metabolites produced in response to stress as

suggested by Keeling and Bohlmann³⁵. Phytochemicals have diverse functions and the presence of alkaloid may be the reason for its use in the treatment of alimentary tract diseases and gastro-intestinal disorders, and also have parasitic and predator repelling effects³⁶, anesthetic and pain killer properties^{37, 38}; Flavonoids have been reported to have anti-viral, anti-allergic, anti-platelet, anti-tumour, antioxidant and anti-inflammatory effect^{36, 39-41}, while tannins inhibit the growth of bacteria, fungi, yeast and viruses⁴² and fight against infection, and are also used as astringents⁴¹; Saponines are useful as expectorants, antibacterial and anti-viral⁴¹; Terpenoids have the properties for prevention and therapy of cancer, antimicrobial, antifungal, antiviral, immunomodulatory and are useful as aromatic agents and flavouring the food^{41, 43}. Preliminary phytochemical screening of plants is very useful for the determination of the active constituents in different solvent extracts.

Table 1: Preliminary phytochemical screening of different parts of *Malachra capitata* in various solvent extracts.

Solvent extracts	Plant parts tested	Phytochemicals tested											
		Alk	Ant	Cou	Fla	Phe	Pro	Qui	Sap	Ste	Suc	Tan	Ter
Petroleum ether	Root	+	-	-	+	+	-	-	+	-	-	-	-
	Stem	+	+	-	+	+	+	-	+	-	+	-	-
	Leaf	+	+	-	+	+	+	-	+	-	+	-	-
Chloroform	Root	-	+	-	-	-	-	-	-	-	-	-	-
	Stem	-	+	+	-	-	-	-	-	-	-	-	-
	Leaf	-	+	+	-	-	-	-	-	-	-	-	-
Ethyl acetate	Root	-	+	+	+	-	-	-	-	-	-	-	-
	Stem	-	+	+	+	-	-	-	-	-	-	-	-
	Leaf	-	+	+	+	-	-	+	-	-	-	-	-
Methanol	Root	+	+	-	-	+	+	-	+	-	-	+	-
	Stem	+	+	-	+	+	+	-	+	-	-	+	-
	Leaf	+	+	-	+	+	+	-	+	-	-	+	-
Ethanol	Root	+	+	-	+	+	+	-	+	-	+	+	-
	Stem	+	+	-	+	+	+	-	+	-	+	+	+
	Leaf	+	+	+	+	+	+	-	+	-	+	+	+
Water	Root	-	+	+	+	-	-	-	-	-	-	-	-
	Stem	-	+	+	+	-	-	-	-	-	-	-	-
	Leaf	+	+	+	+	+	-	-	-	-	-	-	-

Key: Alk: Alkaloids, Ant: Anthocyanoides, Cou: Coumarin, Fla: Flavonoids, Phe: Phenol, Pro: Protein; Qui: Quinone, Sap: Saponin, Ste: Steroids, Suc: Sucrose, Tan: Tannins, Ter: Terpenoids, (+): Present, (-): Absent.

Dry powder samples of different parts of *M. capitata* revealed the presence of alkaloids, anthocyanoides, coumarins, flavonoids, protein, tannin, terpenoids and phenol compounds. The results are presented in the Table 1. From the results it is evident that the ethanol extract of root sample contain more number of compounds (8) as compared to other solvent extracts. Similarly, a maximum of 9 compounds in stem sample and 10 compounds in leaf sample noted in the ethanol extracts as compared to other extracts. Among the compounds tested,

anthocyanoides noted in all plant sample extracts, except in the petroleum ether root extract in which the compound is absent and the next is favonoids. Maximum number of compounds (alkaloids, anthocyanoides, flavonoids, phenols, protein, saponin, sucrose and tannins) was noted by methanol (6 compounds), petroleum ether (4 compounds), ethyl acetate and water (3 compounds) and chloroform (1 compound).

Most of the active principles are found in alcoholic and aqueous extracts of *M. capitata* dry powder samples as compared to other extracts tested in this study. The results revealed the presence of alkaloids, anthocyanoides, coumarins, flavonoids, phenol, protein, tannin and terpenoids compounds in the *M. capitata* samples with variation between samples and extracts. Similar study was also carried out in the aqueous extract of *M. capitata* roots^{22, 44} and recorded the presence of carbohydrates, phenol, flavonoids, glycosides, terpenes, alkaloid, tannins and saponins. These phytochemical contents indicate that the well processed plant parts of *M. capitata* may offer medicinal and chemoprotective benefits to its users.

3.2. Quantitative Phytochemical Analysis: Several studies document the mucilaginous polysaccharide content in the Malvaceae plants and the primary components are composed of rhamnose, galactose, galacturonic acid and glucuronic acid⁴⁵⁻⁴⁸. In this study, various biochemical compounds such as carbohydrates, reducing sugar, starch, protein and lipid were estimated in different parts (root, stem and leaf) of *M. capitata* and the data are presented in Table 2.

Carbohydrate content in different parts of *M. capitata* was ranges from 1.62 ± 0.14 mg/g to 4.02 ± 0.06 mg/g. Maximum carbohydrate content was observed in stem sample (4.02 ± 0.06 mg/g) and is followed by leaf (3.42 ± 0.07 mg/g) and root (1.62 ± 0.14 mg/g). The carbohydrate content in ethanol extract of *M. capitata* is arranged in the descending order: stem > leaf > root.

Reducing sugar content in different parts of *M. capitata* shows variations and it was ranges from 24.6 ± 0.26 mg/g to 41.17 ± 0.21 mg/g. The leaf sample of *M. capitata* had maximum reducing sugar content (41.17 ± 0.21 mg/g) and is followed by root (37.77 ± 0.42 mg/g) and stem (24.6 ± 0.26 mg/g) sample. The reducing sugar content of ethanol extract of *M. capitata* samples was arranged in the following descending order: leaf > root > stem.

Malachra capitata root, stem and leaf samples shows variations in the starch content and it ranges from 8.34 ± 3.67 mg/g to 11.60 ± 5.14 mg/g. The maximum starch content was noted in stem sample (11.60 ± 5.14 mg/g) followed by leaf (11.28 ± 4.99 mg/g) and root (8.34 ± 3.67 mg/g)

sample. The starch content of ethanol extract of *M. capitata* was arranged in the following descending order: stem > leaf > root.

Protein content of different parts of *M. capitata* shows variations and it ranges from $15.42 \pm 0.38 \text{ mg/g}$ to $30.36 \pm 5.76 \text{ mg/g}$. The maximum protein content was noted in leaf sample ($30.36 \pm 5.76 \text{ mg/g}$) followed by root ($22.22 \pm 3.02 \text{ mg/g}$) and stem ($15.42 \pm 0.38 \text{ mg/g}$) sample. The protein content of ethanol extract of *M. capitata* was arranged in the following descending order: leaf > root > stem.

Lipid content of different parts of *M. capitata* shows variations and it ranges from $0.31 \pm 0.03 \text{ mg/g}$ to $0.24 \pm 0.03 \text{ mg/g}$. The maximum lipid content was observed in leaf sample ($0.31 \pm 0.03 \text{ mg/g}$) followed by stem ($0.25 \pm 0.05 \text{ mg/g}$) and root ($0.24 \pm 0.03 \text{ mg/g}$) sample. The lipid content of ethanol extract of *M. capitata* was arranged in the following descending order: leaf > stem > root.

The results of biochemical analysis shows that among the biochemicals analyzed, the maximum amount of reducing sugar ($41.17 \pm 0.21 \text{ mg/g}$), protein ($30.36 \pm 5.76 \text{ mg/g}$) and lipid ($0.31 \pm 0.003 \text{ mg/g}$) in leaf sample of *M. capitata* was noted, while the carbohydrate ($4.02 \pm 0.06 \text{ mg/g}$) and starch ($11.60 \pm 5.14 \text{ mg/g}$) content was maximum in stem sample of *M. capitata* as compared to other samples and molecules tested. The maximum biochemical content in leaf and stem samples of *M. capitata* are found in the following order i.e., reducing sugar > protein > starch > carbohydrate > lipid.

Analysis of phytochemical content in different parts of *M. capitata* ethanol solvent shows more amount of reducing sugar, protein and lipid in leaf sample, while stem sample shows more carbohydrate and starch content than the root sample.

Table 2: Quantification of phytocompounds in the extracts of *Malachra capitata*.

<i>Phytocompounds analyzed</i>	<i>Extracts of different parts of Malachra capitata</i>		
	<i>(mg/g sample)</i>		
	Root	Stem	Leaf
1. Carbohydrate	1.62 ± 0.14^i	4.02 ± 0.06^{hi}	3.42 ± 0.07^i
2. Reducing sugar	37.77 ± 0.42^a	24.60 ± 0.26^{cd}	41.17 ± 0.21^a
3. Starch	8.34 ± 3.67^{gh}	11.60 ± 5.14^{fg}	11.28 ± 4.99^{fg}
4. Protein	22.22 ± 3.02^d	15.42 ± 0.38^{ef}	30.36 ± 5.76^b
5. Lipid	0.24 ± 0.03^i	0.25 ± 0.05^i	0.31 ± 0.03^i

Mean \pm Standard Deviation (n=3); Similar alphabets between rows and columns indicate non-significance at 5% level CD (P=0.05) value (CS =4.47).

Two-way ANOVA Table:

Source	df	SS	MS	F	PROB
TOT	44	8458.685400	192.242850	26.8126	
Trt	14	8243.589297	588.827807	82.1253	0.018 *
Err	30	215.096103	7.169870	1.0000	
C	4	7419.578222	1854.894556	258.7069	0.000 **
S	2	280.926558	140.463279	19.5908	0.001 **
Cs	8	543.084517	67.885565	9.4682	0.002 **
Err	30	215.0961	7.169870	1.0000	
	SED	CD(0.05)	CD(0.01)	CV = 8.90%	
C	1.26226	2.57791	3.47122		
S	0.97774	1.99684	2.68880		
Cs	2.18630	4.46508	6.01233		

*: Significance 5% level;

**: Significance at 1% level.

3.3. Antioxidant compounds: Antioxidants play a vital role in the body defense system against reactive oxygen species (ROS), which are believed as harmful byproducts generated during normal cell aerobic respiration and cause pathological conditions. Total phenol content of different parts (root, stem and leaf) of *M. capitata* was quantified in ethanol solvent and the data are shown in Table 3. Total phenol content in different parts (root, stem and leaf) of *M. capitata* ranges from 9.00mg/g to 190.00mg/g. Maximum phenol content were noted in leaf sample (190.00mg/g) and are followed by stem and root. Dipeki *et al.*²¹ reported the presence of alkaloids, flavonoids, tannin, phlobatanin, terpenoids, saponin and glycosides in aqueous of leaf extract of *Malachra capitata*. Total tannin content of different parts (root, stem and leaf) of *M. capitata* was quantified in ethanol solvent and the data are shown in Table 3. Different parts of *M. capitata* shows variations in total tannin content and it ranges from 0.70mg/g to 11.20mg/g. The leaf sample possesses maximum (11.20mg/g) tannin content when compared with other parts. Vitamin-C content of different parts (root, stem and leaf) of *M. capitata* was quantified in ethanol solvent and the data are shown in Table 3. The amount of vitamin-C present in the different parts of *M. capitata* is found to be maximum 70.18mg/g in leaf sample as compared to root and stem sample.

Table 3: Quantification of antioxidants in the ethanol extracts of *Malachra capitata*.

Plant parts tested	Total phenol (mg/g)	Total tannin (mg/g)	Vitamin – C (mg/g)
1. Root	9.00±02.00 ^{gh}	0.70±0.10 ⁱ	52.63±2.47 ^e
2. Stem	70.00±10.00 ^c	1.41±0.09 ^{hi}	61.40±0.60 ^d
3. Leaf	190.00±10.00 ^a	11.20±0.20 ^{fg}	70.18±2.12 ^{bc}

Mean ± Standard Deviation (n=3); Similar alphabets between rows and columns indicate non-significance at 5% level CD (P=0.05) value (SA =8.10).

Two-way ANOVA Table					
Source	df	SS	MS	F	PROB
TOT	26	85905.138867	3304.043803	150.9807	
Rep	2	79.884800	39.942400	1.8252	
Trt	8	85475.112067	10684.389008	488.2311	0.000 **
Err	16	350.142000	21.883875	1.0000	
S	2	22623.659267	11311.829633	516.9025	0.000 **
A	2	33924.510067	16962.255033	775.1029	0.000 **
Sa	4	28926.942733	7231.735683	330.4596	0.000 **
Err	16	350.142000	21.883875	1.0000	
		SED	CD(0.05)	CD(0.01)	CV =9.85%
S		2.20524	4.67496	6.44128	
A		2.20524	4.67496	6.44128	
Sa		3.81959	8.09727	11.15663	

NS: Non significance; **: Significance at 1% level.

In the present study, *M. capitata* leaf contain maximum amount of phenol, vitamin-C and total tannin content when compared with other parts tested. Deficiency of ascorbic acid is associated with pains in the joint and defect in skeletal calcification, anemia, manifestation of scurvy hemorrhage from mucous membrane of the mouth and gastrointestinal track⁴⁹. This function of ascorbic acid accounts for its demand for normal wound healing. There is also an interesting ability of ascorbic acid as an antioxidant, to prevent or to minimize the formation of carcinogenic substances from dietary material⁴⁹. Kaczmariski *et al.*⁵⁰ reported that among the antioxidative compounds (vitamin -C, -E, -A, selenium and carotenoids), shows very strong intensity of antioxidative activities.

The antioxidant efficacy of phenol compounds is chiefly due to their redox potential and are known to act as reducing agents (free radical terminators), hydrogen donors, metal chelators and singlet oxygen quenchers^{51, 52}. Since it has been shown that phenol compounds of plant kingdom are one of the most effective antioxidative constituents, it is important to estimate the phenolic contents of extracts in order to assess their contribution to antioxidant activity⁵³. Today, there is an increasing interest in tannins as bioactive component of foods as well as biological antioxidants due to its many beneficial properties their ability to integrate strongly with carbohydrates and proteins.

3.4. Anti-microbial activity: Medicinal plants contain components that have therapeutic significance and possess antimicrobial properties. Most important of these bioactive components are alkaloids, flavonoids, tannins, saponins and other phenol compounds⁵⁴⁻⁵⁷.

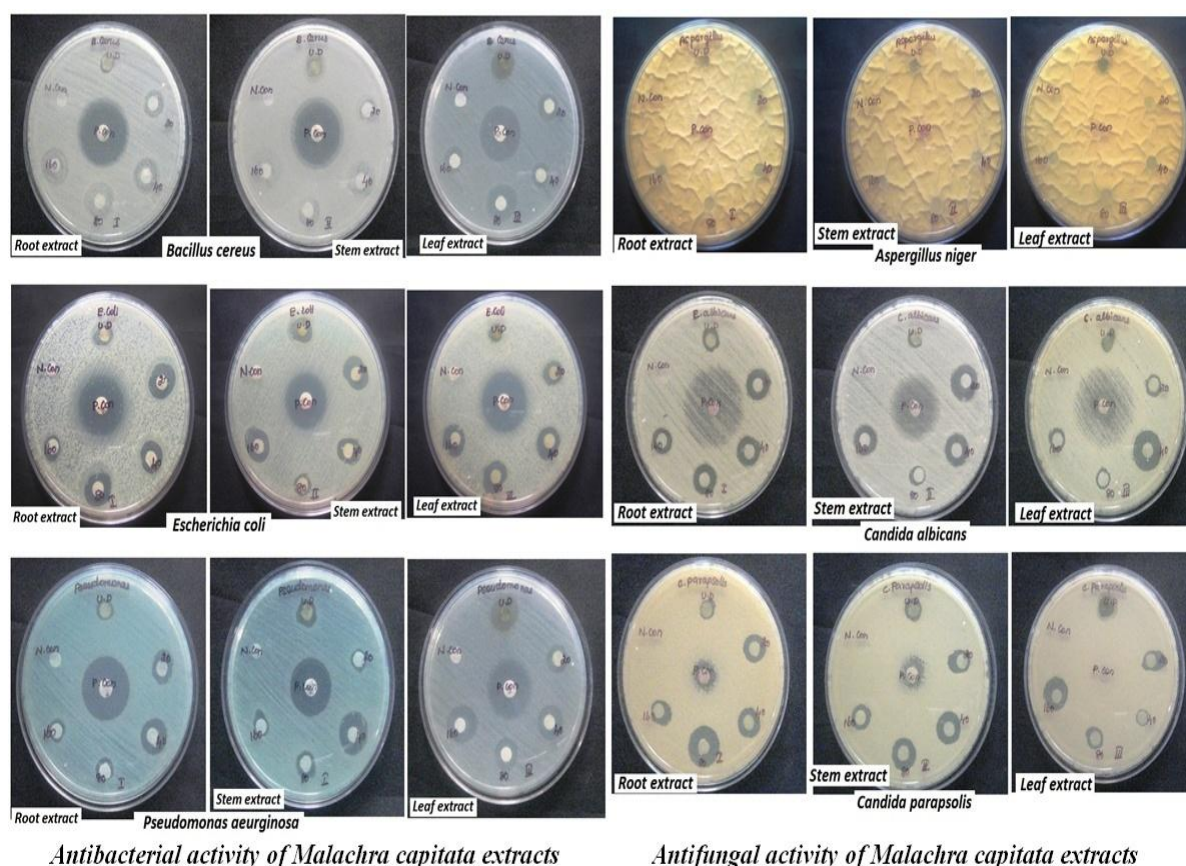


Figure 2: Antimicrobial activity of *Malachra capitata* extracts.

Table 4: Antibacterial activity of *Malachra capitata* extracts.

Test organisms	Plant parts used	Zone of Inhibition (mm)				Netilmycin 30(mcg)	Flucanazole 25(mcg)
		20 (µg/ml)	40 (µg/ml)	80 (µg/ml)	160 (µg/ml)		
1. <i>B. cereus</i>	Root	NR	NR	NR	NR	24.67±1.53	ND
	Stem	NR	NR	NR	NR	22.33±2.08	ND
	Leaf	10.50±0.50	10.93±0.40	15.67±1.04	11.17±1.04	21.67±0.58	ND
2. <i>E. coli</i>	Root	14.50±0.50	11.87±0.81	15.00±1.00	10.00±1.00	20.83±0.76	ND
	Stem	14.67±0.58	11.83±0.76	9.83±0.76	14.67±1.53	20.50±1.32	ND
	Leaf	10.17±1.04	13.80±0.53	13.73±0.64	12.50±0.50	22.67±1.53	ND
3. <i>P. aeruginosa</i>	Root	13.00±0.50	14.17±0.47	10.17±1.3	9.87±0.81	22.33±0.58	ND
	Stem	8.50±0.50	15.97±0.15	9.83±1.76	10.43±1.21	20.67±1.53	ND
	Leaf	11.33±0.58	13.27±0.93	10.83±0.760	15.80±0.72	20.00±1.00	ND

NR: No results under experimental conditions, ND: Not determined. *E. coli*: *Escherishia coli*, *P. aeruginosa*: *Pseudomonas aeruginosa*, *B. cereus*: *Bacillus cereus*, Netilmycin: Positive control for bacteria. Mean ± Standard Deviation (n=3)

Three-way ANOVA Table

Source	df	SS	MS	F	PROB
TOT	161	9692.136111	60.199603	17.8049	
Trt	53	9312.456111	175.706719	51.9679	0.000 **
Rep	2	21.287037	10.643519	3.1480	
Err	106	358.392963	3.381066	1.0000	
O	2	924.001481	462.000741	136.6435	0.000 **
S	2	324.602593	162.301296	48.0030	0.000 **
C	5	5936.535370	1187.307074	351.1636	0.000 **
OS	4	402.990370	100.747593	29.7976	0.000 **
SC	10	226.055926	22.605593	6.6859	0.000 **
OC	10	913.783704	91.378370	27.0265	0.000 **
OSC	20	584.486667	29.224333	8.6435	0.000 **
Err	106	358.392963	3.381066	1.0000	
SED		CD(0.05)	CD(0.01)	C.V. (Trt) =18.68%	
O	0.35387	0.70159	0.92822		

**: Significance at 1% level.

The anti-microbial activity of ethanol extract of *Malachra capitata* (root, stem and leaf) samples against *Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus cereus*, *Candida albicans*, *Candida parapsolisis* and *Aspergillus niger* was studied and the results are presented in Table 4 and 5; Figure 2. Almost all the microbes used in the present study were resistant to the ethanol extract and showed a potential activity against growth of bacterial and fungal organisms.

For anti-bacterial study the microorganisms used were *Escherichia coli*, *Pseudomonas aeruginosa* and *Bacillus cereus*. In the experimental study root and stem sample of *M. capitata* does not possess any activity on *Bacillus cereus*, while the leaf sample shows maximum zone of inhibition 16mm at a concentration of 80µg/ml. In case of *Escherichia coli* maximum inhibition zone of 15mm was observed in root and stem sample at various concentration. In leaf sample maximum zone of inhibition 14mm was found. *Pseudomonas aeruginosa* shows maximum inhibition zone of 16 mm in stem sample at a concentration of 40µg/ml and in leaf at 160µg/ml.

For anti-fungal activity the microorganisms used were *Candida albicans*, *Candida parapsolisis* and *Aspergillus niger*. In *Candida albicans*, maximum inhibition zone of 16mm was reported in leaf sample at a concentration of 40µg/ml followed by 14mm. In *Candida parapsolisis* 17mm inhibition zone was reported in root sample at a concentration of 80µg/ml. But in case of *Aspergillus niger*, *M. capitata* root, stem and leaf sample does not possess any activity.

In the present study, the ethanol extract of different parts of *M. capitata* showed reasonable, comparable inhibitory activity against the growth of bacterial and fungal strains tested. The

bioactives of the extract which elicited antibacterial activity appeared to have preferential and specific activity against *E. coli*, *P. aeruginosa* and leaf sample alone show activity against *B. cereus* where as root and stem sample has no activity against *B. cereus*. The extract also possesses activity against the fungal organisms such as *C. albicans* and *C. parapsolis* since there was no activity against the fungal organism *A. niger*.

Antimicrobial activities have been evaluated with diverse methods often difficult to compare and various reports indicate the potential of pure coumarins against gram-positive and Gram-negative bacteria as well as fungi. The antimicrobial activity of ethanolic leaf extract of *Sida spinosa* Linn against *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Candida albicans*, *Aspergillus niger* showed a potential activity against growth of both Gram positive and Gram negative bacteria and fungus⁵⁸. The petroleum ether and methanol extract of *Abutilon indicum* shows potent antibacterial activity against the pathogenic strains of *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Klebsiella pneumonia*⁵⁹.

Herbal medicine is still the mainstay of about 75-80% of the whole population, and the major part of traditional therapy involves the use of plant extract and their active constituents⁶⁰. The medicinal plants around the world contain many compounds with antibacterial activity⁶¹. The screening of plant extracts and plant products for antimicrobial activity has shown that plants represent a potential source of new anti-infective agents⁶²⁻⁶⁴. The medicinal actions of plants are unique to a particular plant species or group, consistent with the concept that the combination of secondary products in a particular plant is taxonomically distinct⁶⁵. Phenolic compounds (flavonoids, phenolic acids, anthocyanins, stilbenes, tannins, ligans and lignins) are important for normal plant growth and development as well as a defense against infection and injury. These compounds are commonly found in both edible and non-edible plants, and they have been reported to have multiple biological effects, including antioxidant activity⁶⁶. Antibacterial activity of alkaloids from *Sida acuta* is carried out by Daminoti Karou *et al.*⁶⁷ shows that the alkaloids displayed good antimicrobial activity against several test microorganisms.

Table 5: Antifungal activity of *Malachra capitata* extracts.

Test organisms	Plant parts used	Zone of Inhibition (mm)				Netilmycin 30(mcg)	Flucanazole 25(mcg)
		20 (µg/ml)	40 (µg/ml)	80 (µg/ml)	160 (µg/ml)		
<i>I. A. niger</i>	Root	NR	NR	NR	NR	ND	NR
	Stem	NR	NR	NR	NR	ND	NR
	Leaf	NR	NR	NR	NR	ND	NR

2. <i>C. albicans</i>	Root	12.33±0.58	12.67±2.31	11.67±1.53	9.67±1.53	ND	NR
	Stem	14.67±1.15	13.00±1.00	9.33±1.53	11.00±1.00	ND	24.17±1.04
	Leaf	10.00±1.00	14.67±1.15	9.17±1.04	8.67±0.58	ND	27.50±0.50
3. <i>C. parapsolisis</i>	Root	10.67±0.58	11.00±1.00	18.00±1.73	10.33±1.15	ND	12.50±0.50
	Stem	11.00±1.00	13.67±1.15	13.33±1.53	10.67±1.15	ND	12.17±1.04
	Leaf	11.33±1.15	9.33±1.15	10.00±1.00	13.67±0.58	ND	NR

NR: No results under experimental conditions, ND: Not determined, *C. albicans*: *Candida albicans*, *C. parapsolisis*: *Candida parapsolisis*, *A. niger*: *Aspergillus niger*, Flucanazole: Positive control for fungi, Mean ± Standard Deviation (n=3)

Three-way ANOVA Table					
Source	df	SS	MS	F	PROB
TOT	161	8089.766975	50.247000	71.1592	
Trt	53	8013.600309	151.200006	214.1278	0.000 **
Rep	2	1.317901	0.658951	0.9332	
Err	106	74.848765	0.706120	1.0000	
O	2	3559.910494	1779.955247	2520.7531	0.000 **
S	2	53.484568	26.742284	37.8721	0.000 **
C	5	1439.341049	287.868210	407.6758	0.000 **
OS	4	252.719136	63.179784	89.4745	0.000 **
SC	10	311.885802	31.188580	44.1689	0.000 **
OC	10	1152.626543	115.262654	163.2337	0.000 **
OSC	20	1243.632716	62.181636	88.0610	0.000 **
Err	106	74.848765	0.706120	1.0000	
	SED	CD(0.05)	CD(0.01)	C.V. (Trt) =12.74%	
O	0.16172	0.32062	0.42419		

**: Significance at 1% level.

4. CONCLUSION

The results of present study showed that the *Malachra capitata* plant samples tested have some valuable antimicrobial activities due to the presence important primary and secondary metabolites in addition to antioxidants in the plant extracts as reported by many workers.

CONFLICT OF INTERESTS

Authors declare that there is no conflict of interests regarding the publication of this paper.

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