ISSN (ELECTRONIC): 2945-3135

Kempegowda M.S (2026). *Phytochemical Analysis and Antimicrobial activity of Mentha spicata, Mentha piparita, Mentha citrate, Ramanagar Dist, Karnataka. International Journal of Multidisciplinary Research & Reviews, 5(si2), 314-321.*

INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY RESEARCH & REVIEWS

journal homepage: www.ijmrr.online/index.php/home

PHYTOCHEMICAL ANALYSIS AND ANTIMICROBIAL ACTIVITY OF MENTHA SPICATA, MENTHA PIPARITA, MENTHA CITRATE, RAMANAGAR DIST, KARNATAKA

Kempegowda M.S

Department of Botany, Rural Degree College, Kanakapura, Karnataka, India.

How to Cite the Article: Kempegowda M.S (2026). *Phytochemical Analysis and Antimicrobial activity of Mentha spicata, Mentha piparita, Mentha citrate, Ramanagar Dist, Karnataka. International Journal of Multidisciplinary Research & Reviews, 5(si2), 314-321.*

https://doi.org/10.56815/ijmrr.v5si2.2026.314-321

Keywords

Geographical Areas, Crude Aqueous, Microorganisms, Methanol Extraction, Primary Metabolites, Secondary Metabolite, Herbal Practicener, Mentha Piparita, Mentha Citrate, Mentha Spicata.

Abstract

Scientific analysis of plant components follows a logical pathway. Plants are collected either randomly or by following leads supplied by local healers in geographical areas where the plants are found. Initial screenings of plants for possible antimicrobial activities typically begin by using crude aqueous or alcohol extractions and can be followed by various organic extraction methods. Since nearly all of the identified components from plants active against microorganisms are aromatic or saturated organic compounds, they are most often obtained through initial ethanol or methanol extraction. Substances involved in essential metabolic processes within an organism are identified as "primary metabolites". For example lipids, porphyrins, amino acids, polyacids (e.g., citric, tartaric etc.) and many others are among this class. The term "secondary metabolite" is frequently found in literature that addresses the chemistry of plant-derived natural products. The term has no real scientific basis except that over the past half century it has come to represent those products of plant metabolism that are associated with some readily detectable properties (e.g., taste, color, odor), a biological activity In present study three medicinal plants were selected for the antimicrobial activity for the fallowing medicinal plants which has curative properties which is given by the herbal practicener, Mentha piparita, Mentha citrate,

The work is licensed under a Creative Commons Attribution Non Commercial 4.0 International License

ISSN (ELECTRONIC): 2945-3135

Kempegowda M.S (2026). *Phytochemical Analysis and Antimicrobial activity of Mentha spicata, Mentha piparita, Mentha citrate, Ramanagar Dist, Karnataka*. *International Journal of Multidisciplinary Research & Reviews, 5(si2), 314-321.*

	Mentha spicata.

1. INTRODUCTION

It is a well known fact that the plant body is a factory of chemical constituents of different kinds. The phytochemical constituents have played a major role as the basic source for the establishment of several pharmaceutical industries. Studies in phytochemical have lead to the discovery of plant drugs like quinine, morphine, cocaine and reserpine to name a few which have helped in the production of anti- malarial, analgesic. anti-inflammatory and hyposensitive drugs, widely used in medicine today.Many medicinal plants occurring in India are yet to be subjected to rigorous chemical investigation, which may help in discovery of several new drugs. To initiate such investigation phytochemical screening is necessary. Plants have an almost limitless ability to synthesize aromatic substances mainly secondary metabolites, of which at least 12.000 have been isolated, a number estimated to be less than 10% of the total. In many cases, these substances serve as the molecules of plant defense against predation by microorganisms, insects, and herbivores. Further, some of which may involve in plant odour (terpenoids), pigmentation (tannins and quinines), and flavour (capsacin). However, several of these molecules possess medicinal properties. The three samples contain phytochemical compounds like Plavanoids, Terpenes, Steroidal rings, Tanin, Carbohydrates, Terpenoids, Cardiac Glycosides, Alkaloids and Saponin.Indian medicinal plants and their products are used to control diverse diseases such as cataract, bronchitis, pneumonia, ulcers and diarrhoea. Researchers are increasingly turning their attention to folk medicine looking for new leads to develop better drugs against cancer, as well as viral and microbial infections. Although hundreds of plants species have been tested for antimicrobial properties, the vast majority have not yet been adequately evaluated26. Clinical microbiologists have two reasons to be interested in the topic of antimicrobial plant extracts. First, it is very likely that these phytochemicals will find their way into the arsenal of antimicrobial drugs prescribed by physicians; several are already being tested in humans. It is reported that, on an average, two or three antibiotics derived from microorganisms are launched each year. After a downturn in that pace in recent decades, the pace is again quickening as scientists realize that the effective life span of any antibiotic is limited. New sources, especially plant sources, are also being investigated. Second, the public is becoming increasingly aware of problems with the over prescription and misuse of traditional antibiotics. In addition, many people are interested in having more autonomy over their medical care. A multitude of plant compounds (often of unreliable purity) is readily available over-the-counter from herbal suppliers and natural-food stores, and self-medication with these substances is common. It is estimated that there are 250,000 to 500,000 species of plants on Earth. A relatively small percentage (1 to 10%) of these is used as foods by both humans and other animal species. It is possible that even more are used for medicinal purposes.

The work is licensed under a Creative Commons Attribution Non Commercial 4.0 International License

Kempegowda M.S (2026). *Phytochemical Analysis and Antimicrobial activity of Mentha spicata, Mentha piparita, Mentha citrate, Ramanagar Dist, Karnataka***.** *International Journal of Multidisciplinary Research & Reviews, 5(si2), 314-321.*

2. MATERIALS AND METHODS

1. Collection of plant materials:

The medicinal plants will be identified by ethno botanical survey and from traditional healers of Ramanagara, Karnataka and collected for the experimental purpose. The plants and the parts screened, together with their families and vernacular names. Fresh plant material will be washed under running tap water, air dried and then homogenized to fine powder and stored in airtight bottle for further studies. The plants shortlisted for the present investigation are as Artemisia pallens, Ocimum sanctum, Ocimum basilicum

2. Phytochemical analysis:

Extracts will be tested for the presence of alkaloids, tannins, flavonoids, polyphenols and sapnins ete using standard procedures (Kokate 1994). 1:10 dilution of extracts was prepared and then subjected to phytochemical screening of Glycosides, Alkaloids, Steroids and triterpenoid, Flavonoids, Phenols and Saponins.

Tests for Alkaloids:

a) Dragendorffs test : To I ml of test solution, few drops of Dragendorffs reagent was added and observed for orange brown precipitate.

b) Mayor's test : To 1-3 ml of test solution, few drops of Mayer's reagent was added and observed for the precipitate.

c) Hager's test : To 1-3 ml test solution, Hager's reagent was added and observed for yellow precipitate.

d) Wagner's test : To 1-3 ml test solution, few drops of Wagner's reagent was added and observed for reddish brown precipitate.

3. Test for Saponins

a. Foam test: The test solution was shaken and observed for the formation of foam, which has to be stable for at least 15 min.

b. Haemolysis test : 2 ml of 18% sodium chloride was taken in 2 test tubes. To one test tube distilled water and to the other 2 ml test solution was added. A few drops of blood were added to both the test tubes. They were mixed well and observed for haemolysis under the microscope.

Test for Glycosides:

a) Baljet's test: The test solution was treated with sodium picrate and observed for yellow to orange colour.

The work is licensed under a Creative Commons Attribution Non Commercial 4.0 International License

Kempegowda M.S (2026). *Phytochemical Analysis and Antimicrobial activity of Mentha spicata, Mentha piparita, Mentha citrate, Ramanagar Dist, Karnataka. International Journal of Multidisciplinary Research & Reviews, 5(si2), 314-321.*

b) Legal's test (For cardenoloids): To the test solution, Iml pyridine and 1 ml sodium nitroprusside were added and observed for pink to red colour.

c) Test for deoxysugars (Kellar Killani test) To 2 ml test solution, glacialaceticacid, one drop of 5% FeCb and concentrated H2S04 were added and observed for reddish brown colour at the junction of the two liquids and a bluish green upper layer.

d) Test for anthraquinone glycosides

4. Borntrager's test:

The powdered drug was mixed with 5 ml of 10% sulphuric acid for 5 mins. It was filtered while hot. The filtrate was cooled and shaken gently with equal volume of benzene. The benzene layer was separated and then treated half of its volume with solution of ammonia (10%). They were allowed to separate. The ammoniacal layer will acquire rose pink colour ifanthraquinones are present.

5. Tests for Carbohydrates:

Preparation of test solution:

The test solution was prepared by dissolving the test extract with water. Then it was hydrolyzed with 1 volume of2N HC1 and subjected for the following chemical tests.

Barfoed's test:

a) Equal volumes of Barfoed's reagent and test solution were takenanheated for 1-2 minutes in a boiling water bath and cooled. It was then observed for a red precipitate.

b) Fehling's test: 1 ml Fehling's A and Iml Fehling's B solutions were mixed and boiled for one minute. To this solution an equal volume of test solution was added and heated in a boiling water bath for 5-10 min and observed for a brick red precipitate.

Tannins and phenol compounds:

To 2-3 ml extract solution, few drops of following reagents were added:

a) 5% FeC13 solution: and observed for deep blue-black colour.

b) Lead acetate solution: Observed for white precipitate.

c) Bromine water: Observed for the discoloration of bromine water.

d) Acetic acid solution: Observed for red colour.

e) Dilute iodine solution: Observed for a transient red colour.

f) One drop of NH4OH and excess 10% AgNO3 solution were added to the test solution and heated for 20 min in a boiling water bath. White precipitate will be observed and later dark silver mirror deposits on the walls of the test tube.

The work is licensed under a Creative Commons Attribution Non Commercial 4.0 International License

Kempegowda M.S (2026). *Phytochemical Analysis and Antimicrobial activity of Mentha spicata, Mentha piparita, Mentha citrate, Ramanagar Dist, Karnataka*. *International Journal of Multidisciplinary Research & Reviews, 5(si2), 314-321.*

Tests for Proteins:

a) Ninhydrin test: 3 ml of test solution and 3 drops 5% Ninhydrin solution were heated in a boiling water bath for 10 min. It was observed for purple or bluish colour.

b) Million's test: 3 ml of test solution was mixed with 5 ml of Million's reagent, and observed for white precipitate. This precipitate when warmed turns brick red or it will dissolve giving red colour.

c) Biuret test: To 3 ml test solution, 4% NaOH and few drops of 1% CUS04 solution was added and observed for violet or pink colour.

Tests for Steroids:

Preparation of test solution: The extracts were reflux separately with alcoholic solution of potassium hydroxide till complete saponification occurs. The saponified extract was diluted with water and unsaponifiable matter was diluted with diethyl ether. The ethereal extract was evaporated and the residue (unsaponifiable matter) was subjected to the following tests by dissolving the residue in Chloroform;

a) Salkowiski test : To 2 ml of extract solution, 2 ml chloroform and 2 ml concentrated H2S04 were added. They were shaken well, and observed whether chloroform layer appeared red and acid layer showed greenish yellow fluorescence.

b) Libermann-Burchard test : 2ml of the extract was mixed with chloroform and 1-2 ml of acetic anhydride and 2 drops of concentrated H2SC>4 were added to the sides of the test tube. First red, then blue and finally green colour can be observed69.

Antibacterial Activity

Preparation of culture media: Muller Hinton Agar (HIMedia) is used for the antibacterial susceptibility study The ingredients are weighed and added in 1000ml distilled water and boiled to dissolve it completely. The pH of media was adjusted to 7.4 0.2 (at 25 c) and sterilized it by autoclaving at 15 lbs pressure (112C) for 15 min. The solution of the test extracts was prepared at the concentration of 5 mg/ml by dissolving in dimethylsulphoxide (DMSO) in the stopper specific gravity bottle and stored in refrigerator. The solution was removed from the refrigerator one hour prior to use and is allowed to warm up to room temperature.

Antimicrobial Screening of Extracts

Microorganisms: Bacterial and fungal strains were obtained from NCIM. The following Gram-positive, Gram-negative and fungal organisms were used in the present study to determine the antimicrobial activity of the 36 plants extracts.

The work is licensed under a Creative Commons Attribution Non Commercial 4.0 International License

Kempegowda M.S (2026). *Phytochemical Analysis and Antimicrobial activity of Mentha spicata, Mentha piparita, Mentha citrate, Ramanagar Dist, Karnataka. International Journal of Multidisciplinary Research & Reviews, 5(si2), 314-321.*

C. coagulans (NCIM-2313)
B. subtils (NCIM-2063)
S. aureus (NCIM-2079)
Gram negative bacterial organisms
E. Coli (NCIM-2063)
K. pneumonia (NCIM-2249)
P. aeruginosa (NCIM-2200)
Fungal organisms
A flavurs (NCIM-0535)
COLLEJans (NCIM-3471)
Distlemans (NCIM)

The bacterial and fungal stock cultures were maintained on Nutrient agar and Sabouraud-dextrose agar respectively which were stored at 4°C.

A. Preparation of culture media70'71

a) Nutrient Agar Medium

The Nutrient Agar medium was prepared by dissolving 28 g of nutrient agar in 1000 ml of distilled water.

Composition:

Peptone : 1%

NaCI : 0.5%

Beef Extract : 1%

Agar : 2%

pH range : 7.4 ± 0.2

b) Nutrient Broth Medium

The nutrient broth medium was prepared by dissolving 13 g of nutrient broth in 1000 ml of distilled water.

Composition

Peptone : 1%

The work is licensed under a Creative Commons Attribution Non Commercial 4.0 International License

Kempegowda M.S (2026). *Phytochemical Analysis and Antimicrobial activity of Mentha spicata, Mentha piparita, Mentha citrate, Ramanagar Dist, Karnataka. International Journal of Multidisciplinary Research & Reviews, 5(si2), 314-321.*

NaCl	:	0.5%
Beef extract	:	1%
pH range	:	7.4 ± 0.2

The medium was sterilized by autoclaving at 15 lb/sq. inch pressure at 121°C for 20 minutes.

B. Preparation of test inoculum

a) Sub culture (Preparation of seeded broth)

The strains of bacteria were inoculated into conical flasks containing 100 ml of sterile nutrient broth. These conical flasks were incubated at 37°C 1°C for 24 hours. This was stored as seeded broth.

b) Viable count

(i) Dilutions

1 ml of 24 h seeded broth of each strain was diluted with 9 ml of sterile water. 1 ml of this was further diluted to 10 ml with sterile water. This was continued till 10"2 to 10'7 dilutions of the seeded broth were obtained

(ii) Incubation of nutrient agar Petri dishes

The dilution was studied by inoculating 0.1 ml of each dilution on to the solidified nutrient agar medium by spread plate method. After incubation at 37°C 1°C for 24h, the number of well formed colonies on the plates was counted. The seeded broth was then suitably diluted to contain between 10^-10'7 microorganisms per ml or Cfu/ml.

This was designated as the working stock, which was used for antibacterial studies.

Antifungal activity

A) Preparation of culture media

Composition of Sabouraud's Dextrose Agar (SDA) :

Glucose	:	40 g
Peptone	:	10 g
Agar	:	25 g
Distilled water	:	1000ml
Ph	:	5.4

Composition of Sabouraud's Dextrose Broth (SDB)

Glucose	:	40 g
Peptone	:	10g
Distilled water	:	1000ml
PH	:	5.4

The work is licensed under a Creative Commons Attribution Non Commercial 4.0 International License

Kempegowda M.S (2026). *Phytochemical Analysis and Antimicrobial activity of Mentha spicata, Mentha piparita, Mentha citrate, Ramanagar Dist, Karnataka. International Journal of Multidisciplinary Research & Reviews, 5(si2), 314-321.*

3. AUTHOR(S) CONTRIBUTION

The writers affirm that they have no connections to, or engagement with, any group or body that provides financial or non-financial assistance for the topics or resources covered in this manuscript.

4. CONFLICTS OF INTEREST

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

5. PLAGIARISM POLICY

All authors declare that any kind of violation of plagiarism, copyright and ethical matters will take care by all authors. Journal and editors are not liable for aforesaid matters.

6. SOURCES OF FUNDING

The authors received no financial aid to support for the research.

REFERENCES

[1] Ncube NS. Afolayan AJ. Okoh Al. 2008; Assessment techniques of antimicrobial properties of natural compounds of plant origin: current methods and future trends. African Journal of Biotechnology. 7 (12):1797-1806.

[2] Srinivasan D Sangeetha Nathan, Suresh T, Lakshmana Perumalsamy P. 2001 Antimicrobial activity of Certain Indian Medicinal plants used in folkloric medicine. Journal of Ethnopharmacology.;74(3):217-220.

[3] Marjorie Murphy Cowan. 1999 Reviews.: 12(4):564-582. Plant Products as Antimicrobial Agents. Clinical Microbiology

[4] John P. Devlin. 1997 High throughput Screening: the discovery of bioactive substances: p.47-53.

[5] Adekunie AS, 2009 Adekunie OC. Preliminary assessment of antimicrobial properties of aqueous extract of plants against infectious diseases. Biology and Medicine.; 1(3): 20-24.

[6] Gangadevi V. Yogeswari S. Kamalraj S, Rani G, Muthumary J. 2008. The antibacterial activity of Acalypha indica L. Indian Journal of Science and Technology.; 1(6): 1-5

[7] Karthikumar S. Vigneswari K, Jegatheesan K. 2007 Screening of antibacterial and antioxidant activities of leaves of Eclipta prostrata (L). Scientific Research and Essay.; 2(4): 101-104.

[8] Abeysinghe PD, Wanigatunge RP. 2006 Evaluation of antibacterial activity of different mangrove plant extracts. Ruhuna Journal of Science. 1:104-112.

[9] Varaprasad Bobbarala, Prasanth Kumar Katikala. Chandrasekhar Naidu K. Somasekhar Penumajji. 2009 Antifungal activity of selected plant extracts against phytopathogenic fungi Aspergillus niger F2723. Indian Journal of Science and Technology. 2(4):87-90.

The work is licensed under a Creative Commons Attribution Non Commercial 4.0 International License