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Research Article

Antimicrobial and Antioxidant activities of leaf extracts of *Mirabilis viscosa* Cav.

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Abstract:

Plants have been widely used in both medicine and cuisine for centuries. Due to their potent antimicrobial and antioxidant properties, they have gained attention as potential therapeutic agents. In this study, various solvents, including polar and non-polar solvents such as ethyl acetate, acetone, methanol, ethanol, and water, hexane and chloroform were used for extraction. The research focused on analysing bioactive compounds present in *Mirabilis viscosa* leaf extracts and evaluated their antimicrobial and antioxidant properties. The antibacterial and antifungal activities of the leaf extracts were assessed using the agar well diffusion method, along with standard antibiotics, against both gram-positive and gram-negative bacteria as well as fungal species. Among the tested microorganisms, *Staphylococcus aureus* exhibited the highest susceptibility, with inhibition zones measuring 18 ± 0.02 mm, 18 ± 0.01 mm, 16 ± 0.02 mm, and 16 ± 0.01 mm for chloroform, methanol, ethyl acetate, and acetone extracts, respectively. *Candida albicans* demonstrated sensitivity to ethanol extract whereas, *Aspergillus niger* also exhibited sensitivity to methanol extracts, showing an inhibition zone of 12 ± 0.51 mm. To determine antioxidant assays such as DPPH, FRAP, reducing potential, and phosphomolybdate assays were conducted. Among these, the phosphomolybdate assay in the methanol extract exhibited the highest antioxidant activity, measuring 63.98 ± 1.17 mg AAE/g. Further studies on the plant bioactive compounds could lead to their potential use in medicine. Recent findings highlight that *M. viscosa* leaf possess significant antibacterial and antioxidant properties, making them a promising resource for future medicinal applications.

Keywords: Antimicrobial activity, DPPH, FRAP, Phosphomolybdate, Total reducing potential assays and *M. viscosa*.

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1. Introduction

Plants play a vital role in human life, providing essential nutrients for health and nourishment while also offering therapeutic benefits. Various plant forms, including trees, herbs, and leafy vegetables, serve as both food and medicine (Nwaogu *et al.*, 2007). Medicinal plants as a source of antioxidant compounds, producing abundant

antioxidants to combat oxidative stress (Hasani *et al.*, 2007; Ali *et al.*, 2008). Medicinal plants are essential for healthcare, with 80% of the global population relying on them for treatment. Plants play a vital role in treating hepatitis, aging and liver protection (Harman, 1995). Various plant parts, including leaves, roots, bark, fruits, and seeds, possess therapeutic properties (Upadhyay *et*

al., 2011). Their effectiveness is attributed to bioactive compounds like alkaloids, saponins, tannins, flavonoids, and terpenoids, which influence physiological functions in the body (Dahiya and Purkayastha, 2012). Medicinal plant extracts are known for their antibacterial properties, with various studies documenting their bioactive compounds (Daoud *et al.*, 2015). Solvent properties, including polarity, viscosity and vapor pressure influence the extraction of antioxidant compounds affecting their activity (Naczka and Shahidi, 2006; Turkmen *et al.*, 2006). Natural products from higher plants may provide new antibacterial agents with unique mechanisms of action (Ahmad and Aqil, 2007). They not only treat infections effectively but also reduce the side effects commonly associated with synthetic antimicrobials (Iwu *et al.*, 1999). Despite the vast diversity of plant species, only a few have undergone pharmacological or biological screening, leaving much of their medicinal potential unexplored. The antibacterial properties of plants play a key role in their therapeutic effects. Plant-derived antimicrobial compounds could help reduce antibiotic dependence and mitigate the risk of antibiotic resistance in food-borne pathogens (Voon *et al.*, 2012).

Certain plant-derived phytochemicals possess antimicrobial properties, making them valuable candidates for developing new antimicrobial drugs (Nascimento *et al.*, 2000). Investigating the composition and activity of medicinal plants is crucial to validating their potential as sources of antimicrobial compounds (Nair and Chanda, 2006). Due to safety concerns surrounding synthetic antioxidants, there is growing interest in natural, cost-effective alternatives from plants. Polyphenols, the primary antioxidants in plants, exhibit redox properties that enable them to neutralize free radicals, quench reactive oxygen species, and break down peroxides, contributing to their antioxidant activity (Zhang and Wang, 2001). Flavonoids are the most abundant and widely distributed phenolic compounds in plants, known for their ability to neutralize free radicals by scavenging them and inhibiting their production (Kumar *et al.*, 2011). The antioxidant properties of phytochemicals contribute to their effectiveness in treating various diseases (Akinmoladun *et al.*, 2007). In the past, plant secondary metabolites were viewed as anti-nutritional factors in livestock farming. However, with recent limitations on antibiotic growth promoters, there has been growing interest in these bioactive compounds as natural alternatives for enhancing performance (Greathead, 2003). The antioxidant potential of plant extracts can be assessed using standard techniques such as the reducing power assay, DPPH scavenging assay, FRAP and phosphomolybdate assay. This study intends to evaluate the antioxidant potential of various solvent extracts from plant leaves and explore their antimicrobial properties.

2. Materials and Methods

2.1. Collection and authentication

Fresh *Mirabilis viscosa* leaf were collected from field areas in Mysuru. 12.298162° N latitude and 76.637683°

E longitude, with an elevation of around 760 meters. A sample of the plant material was then sent to the Botanical Survey of India (BSI) in Coimbatore, Tamil Nadu, for identification and taxonomic confirmation. The plant was verified by the chairman of Botany, Manasagangothri, University of Mysore and assigned voucher number UOMBOT24MV263. Meanwhile, the collected plants were shade-dried and coarsely ground for further analysis.

2.2. Extraction of plant materials

2.2.1. Solvent Extraction

A Soxhlet apparatus was used to extract 10g of shade-dried leaf powder with 100 ml of various solvents, including water, ethanol, methanol, hexane, chloroform, acetone, and ethyl acetate. The extracts were then dried under reduced pressure using a rotary vacuum evaporator. The resulting dehydrated residue was stored in desiccators until further use (Sasidharan *et al.*, 2011).

2.2.2. Bacterial and Fungal strains

As test organisms, gram-positive and gram-negative bacteria such as *Staphylococcus aureus* MTCC96, *Bacillus subtilis* 168, *E. coli* MTCC118, *Klebsiella pneumoniae* MTCC109, and *Pseudomonas aeruginosa* MTCC424 were employed, along with fungal pathogens such as *Candida albicans* ATCC90028, *Aspergillus niger* MTCC1344, and *Penicillium chrysogenum* MTCC 5108. The cultures of bacteria and fungi were kept at 37°C and 28°C on nutrient agar medium and potato dextrose agar medium respectively.

2.2.3. Preparation of Inoculum

The gram-positive and gram-negative tested bacteria were suspended in double-distilled water and centrifuged at 10,000 rpm for five minutes. Their cell density was then determined using spectrophotometry (A nm). For fungal inoculum preparation, tested fungi were cultured on potato dextrose agar medium for 5 to 10 days. After adding 8 to 10 millilitres of distilled water to the Petri dishes, a sterile spatula was used to scrape the conidia. The spore density of each fungus was then adjusted to a final concentration of 10⁵ spores/ml using a spectrophotometer (A595 nm), following the method described by Vanita *et al.* (2019).

2.3. Evaluation of Antimicrobial Activity by Agar Well Diffusion Assay and Disc Diffusion Assay

M. viscosa leaf extracts were tested for antibacterial and antifungal properties against selected bacterial and fungal strains. Nutrient agar for bacteria and potato dextrose agar for fungi were autoclaved at 121°C for 15 minutes. After cooling, 25 ml of medium was added to Petri dishes and solidified in a laminar air flow chamber. The spread plate technique was used to inoculate bacterial and fungal cultures, followed by the creation of a 4 mm well and disc in the agar. Each well and disc was filled with 80 µl of *M. viscosa* leaf extract (100 mg/ml) and incubated for 2 hours. Gentamycin (1 mg/ml) for bacteria and fluconazole (1 mg/ml) for fungi served as positive controls. Incubation occurred at 37°C for

bacteria and 30°C for fungi. After 24-72 hours, the inhibition zone was measured to assess activity (Bauer *et al.*, 1966; Perez *et al.*, 1990).

2.4. Antioxidant Activities

2.4.1. Total Antioxidant activity.

A mixture of phosphate buffer and 20 µl leaf extract (stock) was prepared to a final volume of 200 µl, followed by the addition of 200 µl of 1% potassium ferricyanide and incubated at 50°C for 20 minutes. The reaction was stopped with 200 µl of 10% TCA, then 0.6 ml distilled water and 94 µl of 0.1% ferric chloride were added. After centrifugation at 8000 rpm for 10 minutes, the supernatant absorbance was measured at 650 nm. The total antioxidant activity was calculated using the equation $y = 0.2067x - 0.3131$ ($R^2 = 0.9477$) based on ascorbic acid standards (10–100 µg) and expressed as ascorbic acid equivalents. (Oyaizu, 1986).

2.4.2. 2,2 – Diphenyl - 1- picryl - hydrazyl (DPPH) free radical Scavenging Assay

20 µl of stock leaf extract was dissolved in 1.98 ml methanol, mixed with 2 ml of 0.16 mM DPPH and incubated for 30 minutes in the dark at 37°C. The control used only the sample buffer. Absorbance at 517 nm was measured using a UV-V spectrophotometer, with methanol as the sample blank. Scavenging activity (%) was calculated using the formula. Scavenging activity = $[(A \text{ control} - A \text{ sample}) / A \text{ control}] \times 100$. DPPH activity was determined using the equation $y = 17.206x - 14.047$ ($R^2 = 0.9992$) based on gallic acid concentrations (10–100 µg) Duan *et al.* (2006).

2.4.3. Ferric Reducing Antioxidant Potential (FRAP) Assay

20 µl of leaf extract was added to 3 ml of FRAP reagent and incubated at 37°C for 30 minutes. Absorbance was measured at 593 nm and the FRAP value was calculated by subtracting the blank absorbance. FRAP activity was expressed as EDTA equivalents and determined using the equation $y = 20.221x - 20.889$ ($R^2 = 0.9899$) based on EDTA concentrations (10–100 µg) (Dudonne *et al.*, 2009).

2.4.4. Phosphomolybdate Assay

20 µl of leaf extract was diluted with 100 µl distilled water and mixed with 1 ml reagent solution (0.6M sulphuric acid, 28 mM sodium phosphate, 4 mM ammonium molybdate). The mixture was incubated at 95°C for 90 minutes, then cooled and measured at 765 nm against the reagent blank. Ascorbic acid was used as a reference. Phosphomolybdate activity was determined using the equation $y = 0.1196x - 0.1003$ ($R^2 = 0.9962$) based on ascorbic acid concentrations (10–100 µg) and expressed as ascorbic acid equivalents (Umamaheswari and Chatterjee, 2008).

2.5. Statistical Analysis

The results of the present study (samples run in triplicates, $n = 3$) are represented as mean values S.D. Analysis of variance was performed and significant differences between mean values were determined by Tukey's test-wise comparison test at a level of significance of $p < 0.05$. Statistical analyses were conducted using SPSS software by one way ANOVA with Geisser- Greenhouse model.

3. Results

3.1. Antibacterial activity

The antibacterial activity of various plant extracts against the tested bacterial pathogens is shown in Table 1. Both gram-positive and gram-negative bacterial strains were found to be susceptible to the extracts. Seven different plant extracts were tested, including water, ethyl acetate, chloroform, acetone, methanol, ethanol, and hexane. Figures 1A to 1E show the zone of inhibition for various bacteria such as *S. aureus* (12–18 mm), *B. subtilis* (12–14 mm), *E. coli* (12 mm), *K. pneumoniae* (12–16 mm), and *P. aeruginosa* (10–14 mm). Antibiotic Gentamycin showed inhibition zones of 22 mm for *S. aureus*, *B. subtilis*, *E. coli*, and *P. aeruginosa*, and 20 mm for *K. pneumoniae*. The aqueous extract showed stronger activity against *E. coli* than other extracts. Gram-positive strains were more susceptible than gram-negative strains. Overall, plant extracts demonstrated moderate antibacterial activity compared to antibiotic Gentamycin, except against *E. coli*.

Table 1. Antibacterial activity of leaves extracts of *M. viscosa* by using Agar well-diffusion assay.

Bacterial Strains	Zone of Inhibition (mm in diameter)							
	Plant extracts							
	Water	Ethyl acetate	Chloroform	Acetone	Methanol	Hexane	Ethanol	Antibiotic
<i>S. aureus</i>	16 ± 0.01	18 ± 0.01	18 ± 0.02	16 ± 0.02	18 ± 0.01	12 ± 0.02	12 ± 0.01	22 ± 0.01
<i>B. subtilis</i>	12 ± 0.01	12 ± 0.02	14 ± 0.03	14 ± 0.02	12 ± 0.01	0 ± 0.00	12 ± 0.01	22 ± 0.00
<i>E. coli</i>	12 ± 0.00	0 ± 0.00	0 ± 0.00	0 ± 0.00	0 ± 0.00	0 ± 0.00	0 ± 0.00	22 ± 0.00
<i>K. pneumoniae</i>	16 ± 0.02	14 ± 0.00	12 ± 0.00	14 ± 0.02	14 ± 0.00	12 ± 0.02	12 ± 0.00	20 ± 0.01
<i>P. aeruginosa</i>	12 ± 0.02	14 ± 0.01	14 ± 0.01	14 ± 0.01	12 ± 0.02	10 ± 0.01	15 ± 0.01	22 ± 0.00

The values are expressed as the mean along with the standard deviation (SD, $n=3$).

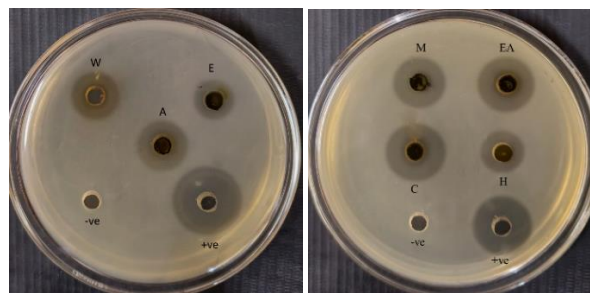


Fig. 1A: Antibacterial activity of leaf extracts against *S. aureus*; A-Antibiotic Gentamycin, -ve: DMSO, (W) Water extract (EA) Ethyl acetate extract (C) Chloroform extract (A) Acetone extract (M) Methanol extract (H) Hexane extract (E) Ethanol extract.

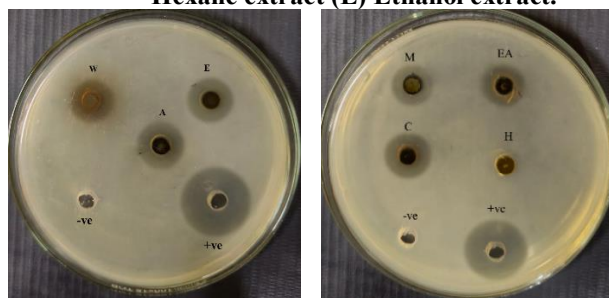


Fig. 1B: Antibacterial activity of leaf extracts against *B. subtilis*; A-Antibiotic Gentamycin, -ve: DMSO, (W) Water extract (EA) Ethyl acetate extract (C) Chloroform extract (A) Acetone extract (M) Methanol extract (H) Hexane extract (E) Ethanol extract.

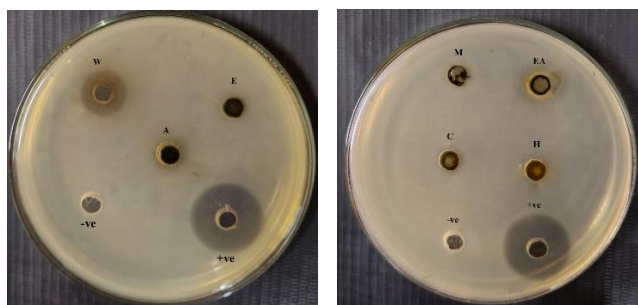


Fig. 1C: Antibacterial activity of leaf extracts against *E. coli*; A-Antibiotic Gentamycin, -ve: DMSO, (W) Water extract (EA) Ethyl acetate extract (C) Chloroform extract (A) Acetone extract (M) Methanol extract (H) Hexane extract (E) Ethanol extract.

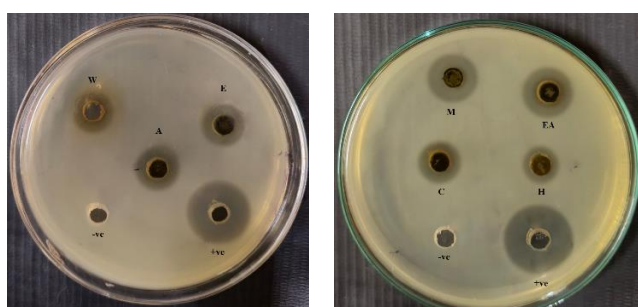


Fig. 1D: Antibacterial activity of leaf extracts against *K. pneumoniae*; A-Antibiotic Gentamycin, -ve: DMSO, (W) Water extract (EA) Ethyl acetate extract (C) Chloroform extract (A) Acetone extract (M) Methanol extract (H) Hexane extract (E) Ethanol extract.

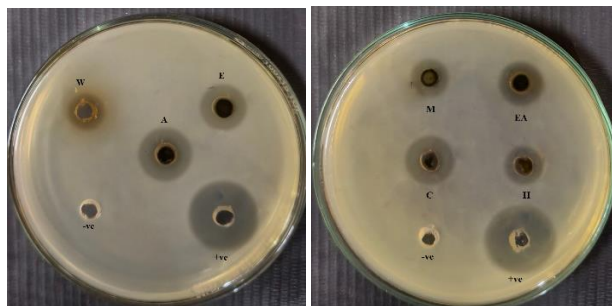


Fig. 1E: Antibacterial activity of leaf extracts against *P. aeruginosa*; A-Antibiotic Gentamycin, -ve: DMSO, (W) Water extract (EA) Ethyl acetate extract (C) Chloroform extract (A) Acetone extract (M) Methanol extract (H) Hexane extract (E) Ethanol extract.

3.2. Antifungal activity

The antifungal properties of different extracts of *M. viscosa* have been assessed in vitro in the current study against the fungus strains *Aspergillus niger*, *Candida albicans* and *Penicillium notatum* which are shown in Table. 2. Ethanol extracts (12 ± 0.51 mm) showed the

largest zone of inhibition in *C. albicans* followed by *A. niger* (12 ± 0.00 mm) in ethanol extract and *P. notatum* (10 ± 0.12 mm) in chloroform extract, compared to all other extracts in figures 2A to 2C. Absence of antifungal activity in the aqueous extract of *A. niger* and *P. notatum*.

Table. 2. Antifungal activity of leaf extracts of *M. viscosa* by using Agar well-diffusion assay.

Fungal Strains	Zone of Inhibition (mm in diameter)							
	Plant extracts							
	Water	Ethyl acetate	Chlorof orm	Acetone	Methanol	Hexane	Ethanol	Antibiotic
<i>C. albicans</i>	10 ± 0.1	10 ± 0.5	10 ± 0.00	10 ± 0.51	10 ± 0.00	8 ± 0.00	12 ± 0.51	16 ± 0.00
<i>A. niger</i>	0 ± 0.00	10 ± 0.12	10 ± 0.11	12 ± 0.11	12 ± 0.51	10 ± 0.00	12 ± 0.00	16 ± 0.00
<i>P. chrysogenum</i>	0 ± 0.00	10 ± 0.7	10 ± 0.12	8 ± 0.00	8 ± 0.11	8 ± 0.05	8 ± 0.12	16 ± 0.01

The values are expressed as the mean along with the standard deviation (SD, n=3).

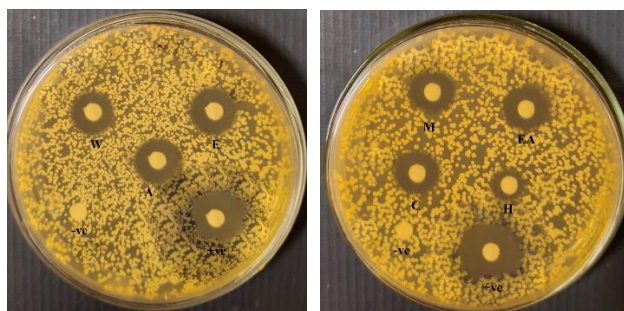


Fig. 2A: Antifungal activity of leaf extracts against *C. albicans*; A-Antibiotic Fluconazole; -ve: DMSO, (W) Water extract (EA) Ethyl acetate extract (C) Chloroform extract (A) Acetone extract (M) Methanol extract (H) Hexane extract (E) Ethanol extract.

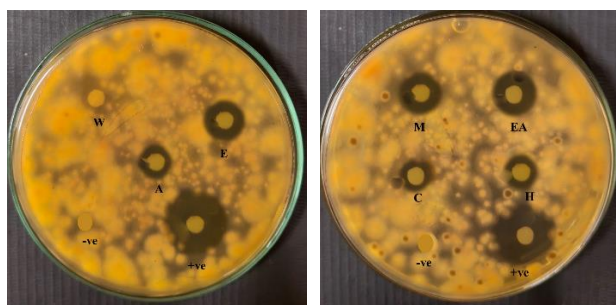


Fig. 2B: Antifungal activity of leaf extracts against *A. niger*; A-Antibiotic Fluconazole; -ve: DMSO, (W) Water extract (EA) Ethyl acetate extract (C) Chloroform extract (A) Acetone extract (M) Methanol extract (H) Hexane extract (E) Ethanol extract.

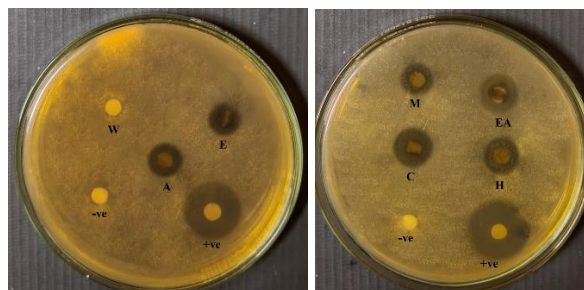


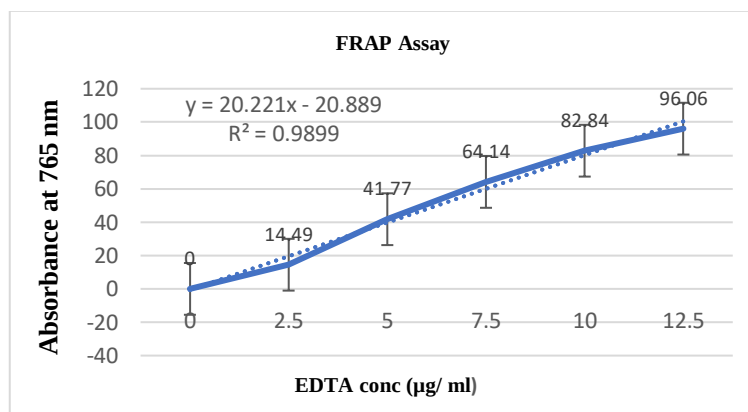
Fig. 2C: Antifungal activity of leaf extracts against *P. notatum*; A-Antibiotic Fluconazole; -ve: DMSO, (W) Water extract (EA) Ethyl acetate extract (C) Chloroform extract (A) Acetone extract (M) Methanol extract (H) Hexane extract (E) Ethanol extract.

3.3. Antioxidant Activities.

3.3.1. FRAP Assay

Our study of various solvent extracts of *M. viscosa* leaf showed antioxidant activity ranging from 2.38 mg EDTAE/g to 4.72 mg EDTAE/g. The methanol extract had the highest FRAP activity (4.72 mg EDTAE/g),

followed by ethanol (4.37 mg EDTAE/g), acetone (3.42 mg EDTAE/g), water (3.34 mg EDTAE/g), hexane (2.94 mg EDTAE/g), ethyl acetate (2.86 mg EDTAE/g), and chloroform (2.38 mg EDTAE/g). FRAP antioxidant capabilities were expressed in EDTA equivalents (mg EDTAE/g) as shown in Table 3 and Graph 1.

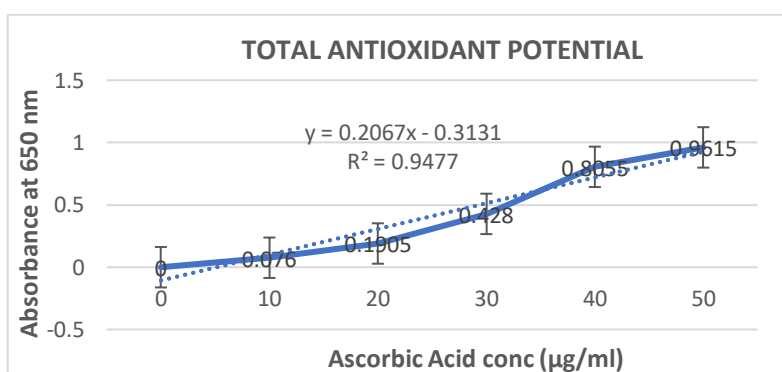


Graph.1 Standard graph of FRAP Assay

3.3.2. Total Antioxidant activity

The methanol extract of *M. viscosa* leaf showed the highest total antioxidant capacity of 47.95 mg AAE/g, followed by ethanol (45.87 mg AAE/g), water (26.69 mg

AAE/g), acetone (24.83 mg AAE/g), ethyl acetate (20.00 mg AAE/g), chloroform (20.00 mg AAE/g), and hexane (19.75 mg AAE/g) as shown in Table 3 and Graph 2.

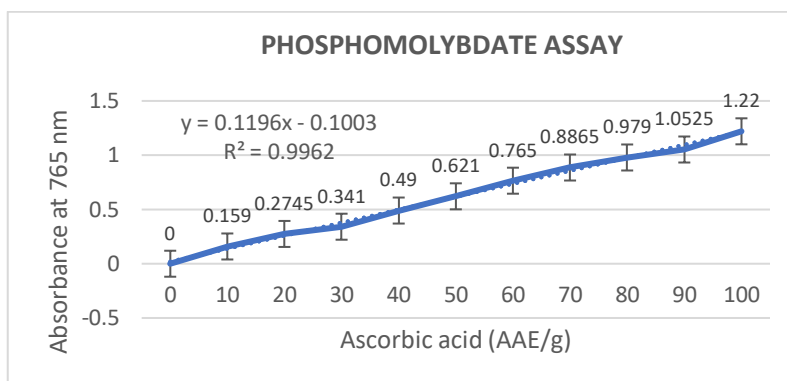


Graph.2 Standard Graph of total antioxidant potential

3.3.3. Phosphomolybdate Assay

The phosphomolybdate activity of various *M. viscosa* leaf extracts is shown in Table. 3 and Graph. 3, with values measured in mg AAE/mg. The methanol extract

exhibited the highest Phosphomolybdate activity (63.98 mg), followed by ethanol (61.35 mg), acetone (52.66 mg), water (39.67 mg), chloroform (35.82 mg), ethyl acetate (27.79 mg), and hexane (26.93 mg).

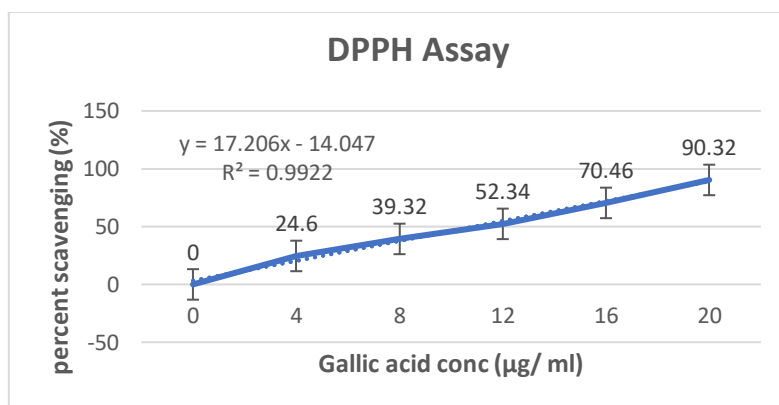


Graph.3. Standard Graph of Phosphomolybdate Assay

3.3.4. DPPH Free radical scavenging Assay

The hexane extract of leaf of *M. viscosa* showed the lowest activity (10.95 mg GAE/g), followed by chloroform (13.30 mg GAE/g), acetone and ethyl acetate (13.58 mg GAE/g), water (15.30 mg GAE/g), ethanol (16.49 mg GAE/g), and methanol (19.82 mg GAE/g). As shown in Table 3 and Graph 4.

M. viscosa half maximum effective concentration (EC₅₀). The EC₅₀ values of *M. viscosa* extracts against DPPH were: hexane 0.98 ± 0.12 mg/ml, chloroform 0.82 mg/ml, acetone, and 0.79 ± 0.21 mg/ml, ethyl acetate 0.77 mg/ml water 0.73 ± 0.01 mg/ml, ethanol 0.65 ± 0.23 mg/ml, and methanol 0.50 ± 0.23 mg/ml.



Graph. 4. Standard Graph of DPPH Assay

Table. 3: Antioxidant activities of different solvent extracts of leaf of *Mirabilis viscosa*

Plant Extracts	FRAP Assay (mg EDTA/g)	Total Antioxidant potential (mgAAE/g)	Phosphomolybdate Assay (mg AAE/g)	DPPH Assay (mg GAE/g)
Water	3.34 ± 0.09 ^c	26.69 ± 1.51 ^b	39.67 ± 0.22 ^c	15.30 ± 1.74 ^c
Methanol	4.72 ± 0.16 ^c	47.95 ± 1.86 ^c	63.98 ± 1.17 ^f	19.20 ± 0.37 ^c
Ethanol	4.37 ± 0.28 ^d	45.87 ± 0.53 ^c	61.35 ± 0.62 ^e	16.48 ± 0.12 ^d
Ethyl acetate	2.86 ± 0.59 ^b	20.00 ± 0.50 ^a	27.79 ± 1.39 ^a	13.58 ± 0.05 ^b
Acetone	3.42 ± 0.70 ^c	24.83 ± 0.55 ^b	52.66 ± 1.10 ^d	13.58 ± 1.86 ^b
Chloroform	2.38 ± 0.88 ^a	20.00 ± 0.05 ^a	35.82 ± 0.8 ^b	13.30 ± 0.50 ^b
Hexane	2.94 ± 0.48 ^b	19.75 ± 1.08 ^a	26.93 ± 0.92 ^a	10.95 ± 0.25 ^a

The values are expressed as the mean along with the standard deviation (SD, n=3).

Discussion

The plant *M. viscosa* leaf extracts were tested for antimicrobial activity against bacterial and fungal strains using the well-diffusion method, with Gentamycin and Fluconazole as controls. The extracts showed varying zones of inhibition. For *M. viscosa*, the inhibition zones ranged from 12 ± 0.00 mm to 18 ± 0.02 mm for bacteria, and 8 ± 0.00 mm to 12 ± 0.51 mm for fungi. Similarly, the plant *M. jalapa* leaf extracts were tested for antimicrobial activity against bacterial and fungal strains. *M. jalapa* inhibition zones ranged from 5 to 15 mm, with the methanol extract demonstrating the largest

inhibition (21 mm) against *S. aureus* and the highest antifungal activity (97.5% at 500 µg/ml) against *A. flavus* (Al- Snafi *et al.*, 2021). The aqueous extract of *M. jalapa* demonstrated strong antimicrobial activity against *P. aeruginosa*, *E. coli*, and *S. aureus* with a zone of inhibition of 11–12 mm (Sumithra *et al.*, 2014). Both studies emphasize the wide-ranging antimicrobial properties of these plant extracts, with *M. viscosa* demonstrating significant antibacterial and antifungal activity.

The antioxidant activity in plant extracts is primarily attributed to phenolic and other antioxidant compounds.

In the FRAP assay, antioxidants reduce the Fe^{3+} /ferricyanide complex to its ferrous form, causing a colour change that is measured by absorbance. An increase in absorbance corresponds to a higher reducing ability as noted by Saeed *et al.* (2012). The methanol extract of *M. viscosa* leaves exhibited the highest FRAP activity (4.72 ± 0.00 mg EDTA/g) demonstrating a strong antioxidant potential. Similarly, *M. jalapa* biogenic nanoparticles showed notable antioxidant activity, reaching 30.85% at 100 $\mu\text{g/ml}$, with an increasing trend as the concentration increased (Pangi *et al.*, 2023). On the other hand, *Pimpinella tragioides* methanol extract displayed an even higher FRAP activity (153.0201 mmol $\text{FeSO}_4/100$ g) (Gohari *et al.*, 2011). These findings suggest that while all three plant extracts exhibit significant antioxidant potential, *P. tragioides* has the highest FRAP activity, followed by *M. jalapa* and *M. viscosa*, confirming the strong antioxidant properties of methanolic extracts across different plant species.

The total antioxidant activity causes the yellow solution to green or blue, depending on the reducing capacity of the compounds. Antioxidants reduce the Fe^{3+} /ferricyanide complex to its ferrous form and the resulting Fe^{2+} is detected by the formation of Perl's Prussian blue, measurable at 700 nm (Ferreira *et al.*, 2007). The methanol leaf extract of *M. viscosa* displayed the highest antioxidant activity among the tested solvents, with a reductional potential of 47.95 ± 0.05 . Similarly, *M. jalapa* exhibited significant antioxidant properties as evidenced by its methanolic extract which demonstrated a reduction potential of 0.4013 ± 0.0015 $\mu\text{g/ml}$ (Zachariah *et al.*, 2012). Moreover, *M. jalapa* nanoparticles showed an even greater total reduction potential (90.89 ± 7.6 AAE/g) in a study by Nadeem *et al.* (2019). Additionally, the ethyl acetate fraction (ESF) of *M. jalapa* flowers exhibited the highest total reducing potential (480.90 ± 0.59 mg AAE/g of dry extract) (Harun *et al.*, 2023). These results indicate that both *M. viscosa* and *M. jalapa* possess strong antioxidant properties, with their methanol extracts displaying considerable reductive potential. However, *M. jalapa* nanoparticles and specific fractions further enhance its total antioxidant activity, while *M. viscosa* shows comparable strong antioxidant effects through its methanol extract.

Total antioxidant capacity (TAC) assays use hydrogen atom transfer (HAT) and single electron transfer (SET) with SET being simpler due to its colour change mechanism. This study assessed the extracts reduction potential using a SET-based phosphomolybdate method, where Mo^{6+} is reduced to Mo^{5+} forming a green phosphate/ Mo^{5+} complex in an acidic medium (Ahmed *et al.*, 2015). The methanol extract of *M. viscosa* exhibited the highest phosphomolybdate activity at 63.98 ± 0.02 mg AAE/g, while the ethanol extract of *B. variegata* demonstrated a greater phosphomolybdate activity of 257.5 μg PGE/g (Mishra *et al.*, 2011). These findings indicate that both plant species possess strong antioxidant potential, though their activity levels vary depending on the extraction solvent used. While *B. variegata* shows higher phosphomolybdate activity, *M.*

viscosa also exhibits significant antioxidant capacity through its methanol extract, highlighting the effectiveness of both solvents in extracting bioactive compounds that contribute to antioxidant activity.

The DPPH assay measures the reactivity of the substances with a stable free radical. At 517 nm, DPPH exhibits strong absorbance, which decreases when antioxidants neutralize the radical causing a colour shift from deep violet to light yellow. The reduction in absorbance reflects the extracts radical-scavenging capacity (Rao *et al.*, 2010). The DPPH assay results indicated strong antioxidant activity in *M. viscosa* leaf extracts with the methanol extract exhibiting the highest DPPH activity at 19.82 ± 0.02 mg GAE/g. Comparatively *M. buxifolia* demonstrated a slightly higher DPPH radical scavenging activity of 24.1 ± 1.02 (Jan *et al.*, 2010), while *M. jalapa* also showed notable antioxidant potential, with its methanolic root extract recording an activity of 17.54 ± 0.10 (Zachariah *et al.*, 2011). Although *M. buxifolia* displayed a higher DPPH activity, *M. viscosa* exhibited superior antioxidant potential compared to *M. jalapa*, reinforcing the effectiveness of its methanol extract. These findings suggest that while different plant species exhibit varying antioxidant capacities *M. viscosa* demonstrates a strong and reliable activity making it a promising source of natural antioxidants.

Therefore, the antibacterial and antioxidant properties of *M. viscosa* were evaluated, confirming its potential as a cost-effective and safe source of natural antioxidants for health supplements.

Conclusion

Our findings support the medicinal use of *M. viscosa* demonstrating its broad-spectrum antimicrobial activity and strong free radical scavenging potential. This study highlights its potential as a natural source of antimicrobial compounds, marking the first report on its mechanism in inhibiting bacterial and fungal growth. The varying phenolic content influences its antioxidant and radical scavenging properties suggesting its role in managing oxidative stress-related illnesses. These results emphasize *M. viscosa* as a promising source of natural antioxidants and antibacterial agents, encouraging further research and clinical trials to explore its potential in developing novel medicines and nutraceuticals.

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Data availability

Non database were generated or analysed during the current study

Declaration

Competing interests

The authors declare no competing interests.

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