Research Article

Bioactive Metabolites and Anti-Phytopathogenic Potential of *Erica verticillata*: A Targeted Leaf Extract Study

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Abstract | Plant pathogenic microorganisms pose significant threats to agriculture, necessitating the exploration of natural alternatives to conventional pesticides. This study aimed to investigate the antimicrobial potential of methanolic extracts from *Erica verticillata* against various plant pathogenic bacteria and fungi. The antibacterial assays revealed substantial activity of the extract, indicating that *Pectobacterium atrosepticum* and *Streptomyces scabiei* exhibited sensitivity at a concentration of 300 µg/ mL. At the same time, *Ralstonia solanacearum* and *Pectobacterium carotovorum* displayed sensitivity at a concentration of 500 µg/ mL. Antifungal evaluations revealed potent inhibition against *Fusarium oxysporum*, *Botrytis cinerea*, and *Rhizoctonia solani*. The extract strongly inhibited *B. cinerea* (78.2 % at 5000 µg/ mL) outperforming copper hydroxide (58.7 %). Significant *in vitro* inhibition was also observed against *R. solani* and *F. oxysporum*, often exceeding the positive control. Phytochemical analysis using HPLC revealed that the predominant phenolic components were gallic acid, caffeic acid, and syringic acid, with concentrations of 2620.3, 606.5, and 580.9 µg/ g, respectively. The principal flavonoid was quercetin at a concentration of 169.4 µg/ g. Hexadecanoic acid and phorbol were the most prevalent secondary metabolites identified in the GC-MS analysis, comprising 26.4 % and 16.4 %, respectively. These findings highlight the rich phytochemical profile of *E. verticillata* and underscore its potential as a source of natural antimicrobial agents for managing plant diseases, including those caused by antibiotic-resistant pathogens.

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Introduction

Plant pathogenic microorganisms, encompassing bacterial and fungal diseases, represent a substantial risk to agricultural production and global food security (Aamer *et al.*, 2024). Bacterial and fungal pathogens, including *Pectobacterium carotovorum*, *Pectobacterium atrosepticum*, *Ralstonia solanacearum*, *Streptomyces scabiei*, *Fusarium oxysporum*, *Botrytis cinerea*, and *Rhizoctonia solani*, are implicated in numerous plant diseases that impact crop quality and yield (Erbs and Newman, 2024; Ali *et al.*, 2025). The increasing resistance of such pathogens to conventional antimicrobial agents necessitates the development of alternative control strategies. The emergence of antimicrobial resistance has become a global health concern, prompting researchers to explore alternative sources of antimicrobial agents (Cotugno *et al.*, 2025). Plant-derived compounds have garnered significant attention due to their diverse bioactive constituents and potential therapeutic applications (Pereira *et al.*, 2025). Medicinal plants have been utilized for centuries in traditional medicine systems worldwide, serving as a valuable reservoir of novel antimicrobial compounds with diverse mechanisms of action (Bhattacharjee and Sharma, 2025).

Recent studies demonstrated the efficacy of plant-derived antimicrobial agents against various plant pathogens (Košćak *et al.*, 2025; Pérez-Flores *et al.*, 2025). The significant antibacterial activity of the phenolic-rich plant extracts against the phytopathogenic bacteria, such as *R. solanacearum* has been reported, highlighting their potential as eco-friendly alternatives to the synthetic bactericides (Yihune and Yemata, 2019). Similarly, the antifungal properties of the plant extracts against phytopathogenic fungi emphasize their applications in sustainable agriculture and forestry (Nguyen *et al.*, 2024). The growing body of evidence supporting the antimicrobial efficacy of plant extracts underscores their potential as natural alternatives to conventional antimicrobial agents in agricultural and pharmaceutical applications (Chaachouay and Zidane, 2024). Despite the promising antimicrobial properties of various plant extracts, comprehensive studies investigating the relationship between their phytochemical composition and biological activities remain limited (Kumar *et al.*, 2023; Murugan *et al.*, 2025). Understanding this relationship is essential for developing standardized plant-based antimicrobial formulations with consistent efficacy and safety profiles. Moreover, identifying specific bioactive compounds responsible for the observed antimicrobial activities could facilitate the development of novel antimicrobial agents with enhanced potency and selectivity.

Plant secondary metabolites, particularly phenolic compounds, have been extensively studied for their antimicrobial potencies (Benalach *et al.*, 2025). These compounds exhibit diverse mechanisms of action against microbial pathogens, including disruption of cell membranes, leading to increased permeability and subsequent cell death, inhibition of microbial enzymes, and suppression of virulence factors (Arzani *et al.*, 2025; De Rossi *et al.*, 2025). A recent study highlighted the significant correlation between the phenolic content of plant extracts and their antimicrobial efficacy against various pathogenic microorganisms (Pérez-Flores *et al.*, 2025). Identifying and characterizing the bioactive compounds in the plant extracts are crucial for understanding their antimicrobial mechanisms and potential applications. Advanced analytical techniques such as high-performance liquid chromatography (HPLC) and gas chromatography-mass spectrometry (GC-MS) have revolutionized the field of natural product studies, enabling the precise identification and quantification of bioactive constituents in the complex plant matrices (Debnath *et al.*, 2025). HPLC analysis is particularly effective for separating and quantifying phenolic compounds, while GC-MS provides valuable information about the volatile and semi-volatile components of plant extracts.

The genus *Erica* (family *Ericaceae*) comprises approximately 860 species distributed across Europe, the Mediterranean region, and South Africa (Dias *et al.*, 2015). Various *Erica* species have demonstrated significant biological activities, including antimicrobial, antioxidant, and anti-inflammatory properties (Guendouze-Bouchefa *et al.*, 2015). *Erica verticillata*, a species native to the Cape Floristic region of South Africa, has been traditionally used for treating various ailments, including respiratory infections and urinary tract disorders (Ojewole, 2008). Despite its ethnomedicinal significance, comprehensive scientific investigations into the antimicrobial potential and phytochemical composition of *E. verticillata* remain limited. The objectives of this study were to evaluate the antibacterial and antifungal activities of *E.*

verticillata methanolic extract against selected plant pathogenic microorganisms, and correlate these activities with the phytochemical composition of the extract. This study employs a multifaceted approach, combining *in vitro* antimicrobial assays with advanced analytical techniques (*i.e.*, HPLC and GC-MS) to provide comprehensive insights into the antimicrobial potential and chemical profile of *E. verticillata* extract. The findings of this study could contribute to the development of natural antimicrobial agents for sustainable plant disease management and potentially address the growing challenge of antimicrobial resistance in agricultural settings.

Materials and Methods

Source of bacterial and fungal strains

This study utilized various bacterial and fungal strains to evaluate the antimicrobial activity of a methanolic extract obtained from leaves of *E. verticillata*. The bacterial strains comprised *P. carotovorum* (GenBank accession number OQ878656), *P. atrosepticum* (GenBank accession number MG706146), *R. solanacearum* (GenBank accession number OQ878653), and *S. scabiei* (GenBank accession number OR437480). These strains were previously isolated from potato plants exhibiting characteristic symptoms of bacterial infections. The fungal strains, including *F. oxysporum* (GenBank accession number OQ820156) sourced from infected potato tubers, *B. cinerea* (GenBank accession number MN398400) obtained from infected strawberry fruits, and *R. solani* (GenBank accession number OQ880457) isolated from infected bean plants.

Preparation of a plant extract

Healthy plant samples of *E. verticillata* were collected from Jebel Akhdar in Libya. Plants devoid of visible morphological diseases were selected. The plant materials were transported to the laboratory, where they were washed with tap water to remove debris and contaminants. The washed plant materials were dried at room temperature (25±2 ˚C) in the shade for 10 d until fully desiccated, a method recommended for preserving biologically active phenolic compounds (Dai and Mumper, 2010). Dried samples were ground using a mill to obtain particles of 0.5 mm, and 50 g of the powder were extracted with 500 mL of 80 % methanol in a rotary shaker at 100 rpm for 12 h at room temperature. This method is deemed adequate for extraction of phenolic compounds from plant materials, as evidenced by prior studies conducted by Azwanida (2015), Altemimi *et al.* (2017). The supernatant; free of plant debris, was obtained *via* filtration using Whatman No. 1 filter paper, and finally the methanol was evaporated at 25-30 ˚C in a rotary evaporator Re-52A (Wincom, Jiangsu, China).

In-vitro antibacterial assay

The agar disc diffusion method was used to evaluate the antibacterial potential of the methanolic plant extract (Bashir *et al.*, 2024). Briefly, a single colony of the purified bacterial strain was inoculated into 100 mL of the nutrient broth (NB) medium and incubated overnight at 27 ˚C. At the end of the incubation period, bacterial growth concentrations were adjusted to 10^8 CFU/ mL using NB medium. Afterward, 100 µL were transferred and spread on the surface of Glycerol Nutrient Agar (GNA) using a sterile glass spreader. Different extract concentrations were prepared in 10 % DMSO to final concentrations of 300, 500, 700, and 1000 µg/ mL in 100 mL of NB medium. The negative control group of the experiment was adjusted using 10 % DMSO in NB medium without the extract. Aliquots of 15 µL of each extract concentration were added to 5 mm diameter filter paper discs, and then left to dry at 4 ˚C for 24 h. The plant extract-loaded filter paper discs were placed individually on surface of the seeded plates and incubated at 27 ˚C for 24 h. The positive control groups were adjusted using amoxicillin as a standard antibiotic with a concentration of 25 µg/ disc. After incubation, the antibacterial activities were quantified by measuring the inhibition zone diameter (mm) using a calibrated ruler in triplicate against the control groups.

The inhibition percentages (%) were calculated based on the obtained inhibition zone diameters using the following formula:

$$Inhibition\ percentage\ (\%) = \left(\frac{Treatment\ inhibition\ zone\ diameter}{Amoxicillin\ inhibition\ zone\ diameter}\right) \times 100$$

(Valgas *et al.*, 2007).

In-vitro antifungal assay

The antifungal effects of the plant extract against some plant pathogenic fungi were evaluated using the radial growth assay on poisoned food procedures, as previously described by Khalil *et al.* (2024). Briefly, different extract concentrations were added to the potato dextrose agar (PDA) plates to obtain final concentrations of 1000, 2000, 3000, 4000, and 5000

µg/mL. A 5 mm disc of 5-day-old fungal culture was placed at the midpoint of each treated plate. The concentration of each extract was evaluated against the negative control (DMSO-PDA) and the positive control (Copper hydroxide) at a concentration of 1.5 g/ L. All plates were incubated for 7 d at 25 ˚C. After incubation, the antifungal activities were detected by determining the fungal mycelial growth inhibition percentage, which was calculated according to the following equation:

Mycelial growth inhibition (%) = [(A0 - At)/A0] × 100

Where A0 is the average diameter in gth untreated plates (control), and At is the average diameter of fungal growth in the treated plates (Bashir *et al*., 2024).

High-performance liquid chromatography analysis

Polyphenolic compounds were identified in the methanol extract using high-performance liquid chromatography (HPLC) (Agilent 1260 Infinity HPLC Series, Santa Clara, CA, USA), which was outfitted with a quaternary pump. The conditions for HPLC were established as follows: A Zorbax Eclipse Plus C18 column (100 mm × 4.6 mm) from Agilent Technologies (Santa Clara, CA, USA), was utilized for separation at 30 ˚C. The separation process was carried out with a 20 µL injection volume, employing a ternary linear elution gradient of water with 0.2 % H_3PO_4 (HPLC grade v/v), methanol, and acetonitrile. The detected peaks were analyzed with a VWD detector calibrated to 284 nm, utilizing a range of phenolic compound standards, including gallic acid, catechol, *p*-hydroxybenzoic acid, caffeine, vanillic acid, caffeic acid, syringic acid, vanillin, *p*-coumaric acid, ferulic acid, ellagic acid, benzoic acid, o-coumaric acid, salicylic acid, and cinnamic acid. Analysis using HPLC is regarded as a highly accurate and trustfull technique for identifying and examining the polyphenolic compounds in the extract, as supported by several prior studies (Naczk and Shahidi, 2004; Abdelkhalek *et al*., 2024).

Gas chromatography-mass spectrometry analysis

Gas chromatography-mass spectrometry (GC-MS) analysis is a reliable technique for identifying volatile and semi-volatile organic compounds in plant extracts. The potential bioactive ingredients in the extract were determined using an (Agilent Technologies, Santa Clara, Ca, USA),, which included an Agilent mass spectrometric detector featuring a direct capillary interface and a fused silica capillary column (HP-5MS), with specifications of 30 m × 0.32 mm × 0.25 µm film thickness. The column temperature was initially set at 50 ˚C with a ramp rate of 5 ˚C/min and then increased to 230 ˚C. The temperature was maintained at 290 ˚C for 2 min. before being increased to the final temperature of 300 ˚C. The sample injection process and program conditions were executed according to a previously published protocol (Youssef *et al*., 2021). The bioactive components of the extract were identified through a search of mass spectral libraries (NIST and Wiley) and by comparing the mass spectra and retention times with information available in the Wiley and NIST mass spectral library databases.

Statistical analysis

A one-way analysis of variance (ANOVA) was performed using the CoStat program (version 6.45) from the Cohort program to analyze the data statistically. The Tukey honest significant difference (HSD) test was used to evaluate differences and determine the statistical significance of the procedure at a significance level of $p \leq 0.05$. An average value (mean ± standard deviation [SD]) together with its standard deviation was displayed for the data. The letters arranged in descending order demonstrated the link and indicated statistical significance (where; a > b > c). The comparable letters indicated no significant alteration.

Results

Antibacterial activity of Erica verticillata extract

Figure 1 illustrates the antibacterial potency of the methanolic extract of *E. verticillata*. The data indicated different sensitivity levels among the tested bacterial strains in response to the extract treatments. *P. atrosepticum* and *S. scabiei* exhibited the highest sensitivity, even at reduced treatment concentrations (300 µg/ mL), recording inhibition percentages of 33.33 % and 62.48 %, respectively.. The results indicate that these bacterial strains exhibited a notable vulnerability to the bioactive compounds present in the extract. *R. solanacearum* and *P. carotovorum* showed moderate sensitivity, with inhibition percentages of 60.51 % and 28.36 %, respectively, when treated with 700 µg/ mL of the extract. This evidence suggests that larger amounts of the extract were necessary to suppress the growth of these bacterial strains effectively. Whereas, *R. solanacearum* and *S. scabiei*

bacterial strains demonstrated a notable sensitivity to the plant extract treatments at a concentration of 1000 µg/ mL, resulting in inhibition percentages of 63.19 % and 75.05 %, respectively. This discovery highlights the potential of *E. verticillata* extract as a viable antimicrobial option for combating multi-drug resistant bacteria. The statistical analysis revealed no significant differences in in the inhibition percentages across all the tested concentrations of the plant extract against the bacterial strains ($p < 0.05$). This finding shows that the antibacterial impact of the methanolic extract reaches an inhibitory limit at a specific concentration; however, adding more concentration does not significantly enhances its ability to inhibit the bacteria.

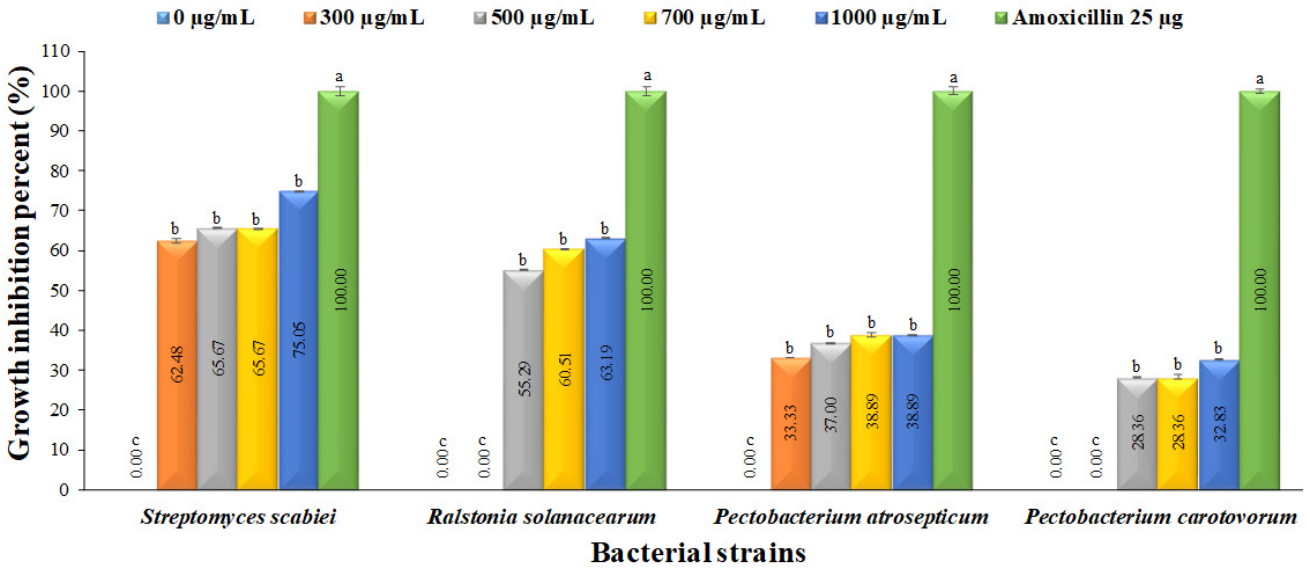


Figure 1: *Antibacterial efficacy of varying doses of E. verticillata methanolic plant extract against the tested bacterial strains, displayed via percentage (%) of growth inhibition. The standard deviation value (±SD) determined for three replicates is represented by error bars. Tukey's HSD test, using a significance threshold of 0.05, indicated that values within each column designated by the same letter (a, b, and c) do not demonstrate statistically significant differences.*

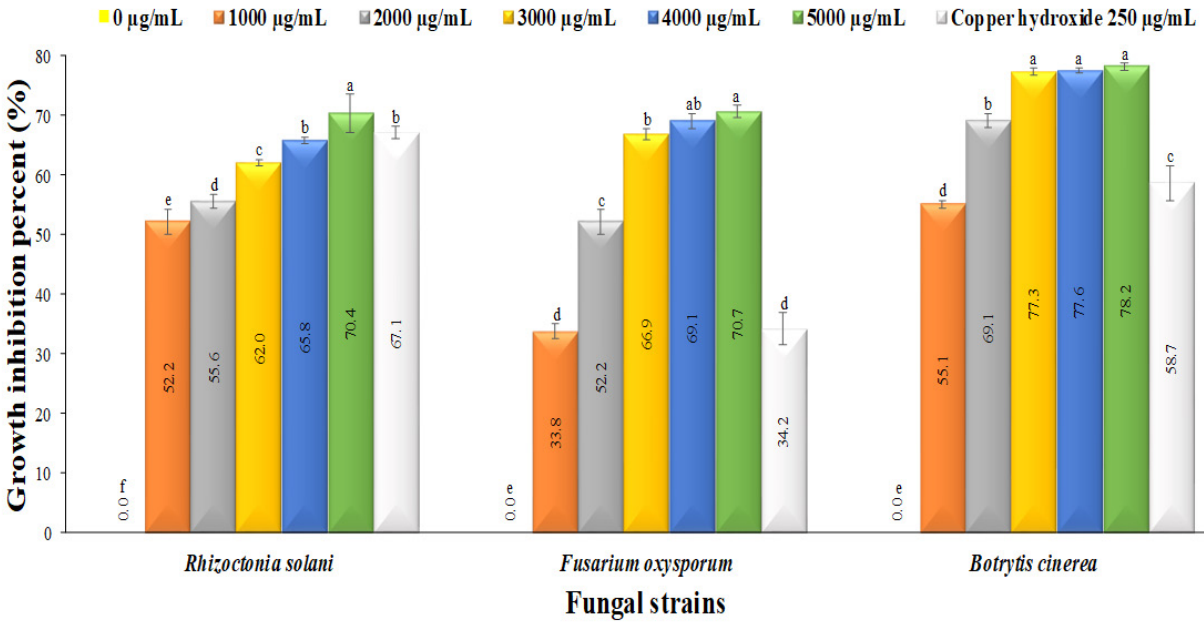


Figure 2: *Antifungal properties of the methanolic extract of E. verticillata against the three tested phytopathogenic strains. The activity was quantified as a percentage (%) of growth inhibition. Each column denotes the mean value of five replicates, and the error bars signify the standard deviation (±SD). Tukey's HSD test, using a significance threshold of 0.05, indicates that values within each column designated by the same letter (a, b, c, d, and e) do not demonstrate statistically significant differences.*

Antifungal activity of Erica verticillata extract

The antifungal properties of methanolic extract from *E. verticillata* were evaluated against three significant plant pathogenic fungi: *F. oxysporum*, *B. cinerea*, and *R. solani*. The findings are depicted in Figure 2. The extract had substantial inhibitory effects against all the tested fungal strains, particularly demonstrating notable action against *B. cinerea*. At 5000 µg/ mL, the extract displayed an impressive fungal growth inhibition rate of 78.2 % against *B. cinerea*, notably surpassing the positive control (copper hydroxide 1.5 g/L), which exhibited only 58.7 % inhibition. Statistical analysis revealed no significant differences ($p < 0.05$) in the efficacy of the extract against *B. cinerea* when administered at concentrations of 3000, 4000, and 5000 µg/ mL. These findings suggest that a lower concentration (3000 µg/mL) effectively achieved maximum inhibitory potential against this fungal pathogen, presenting a cost-effective alternative for potential applications. The extract demonstrated significant activity ($p < 0.05$) against *R. solani* at a concentration of 5000 µg/mL, with an inhibition percentage of 70.4 %.

The obtained results revealed a performance level of the extract comparable to that of the copper hydroxide treatment (67.1 %) and displayed a significant improvement over the untreated control. The obtained data suggest that the extract may be a natural substitute for chemical fungicides in managing *R. solani* infections. Similarly, notable suppression of *F. oxysporum* growth was recorded after applying plant extract concentrations of 2000, 3000, 4000, and 5000 µg/ mL, yielding inhibition percentages of 52.2 %, 66.9 %, 69.1 %, and 70.7 %, respectively. The values observed were significantly greater than the inhibition obtained through copper hydroxide treatment (34.2 %) at $p < 0.05$, highlighting the enhanced antifungal properties of the *E. verticillata* extract compared to the traditional fungicides.

High-performance liquid chromatography (HPLC) analysis for the detection of phenolic and flavonoid compounds

The chemical composition of *E. verticillata* methanolic extract was analyzed using HPLC to identify and quantify the phenolic and flavonoid compounds present. Figure 3 and Table 1 display the HPLC chromatograms and the obtained quantitative results. The HPLC analysis indicated that gallic acid

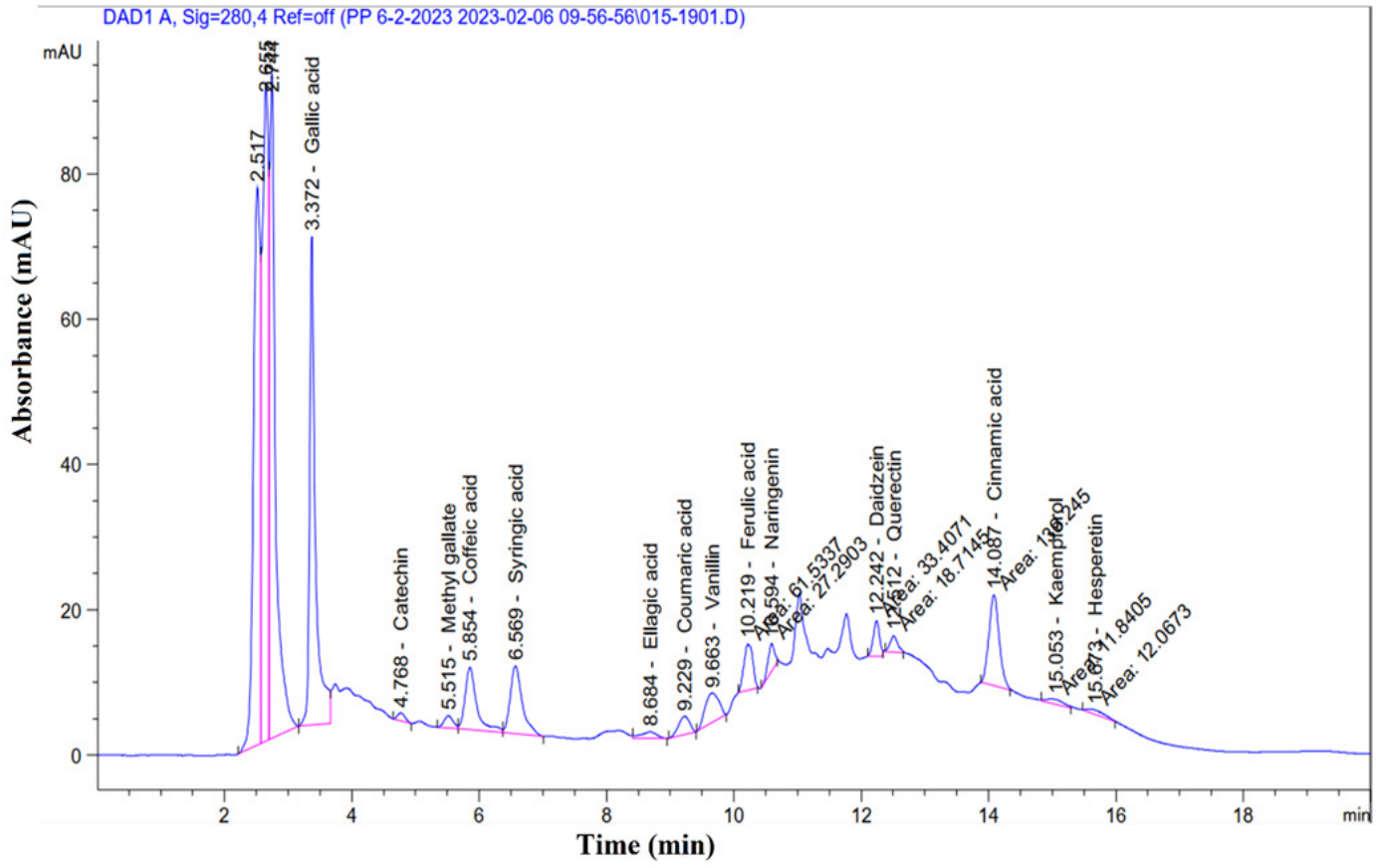


Figure 3: *High-performance liquid chromatography chromatogram of the detected phenolic and flavonoid bioactive compounds in Erica verticillata methanolic extract*.

(2620.3 µg/ g), caffeic acid (606.5 µg/ g), and syringic acid (580.9 µg/ g) were the predominant phenolic compounds in the methanolic plant extract. The potent antioxidant and antimicrobial characteristics of these phenolic acids may substantially enhance the antibacterial and antifungal effects observed in the extract. Naringenin (198.4 µg/ g) and quercetin (169.4 µg/ g) were identified as the most abundant flavonoid compounds in the extract. Flavonoids exhibit a range of biological activities such as antimicrobial effects, and their inclusion in the extract likely enhances its overall bioactivity. Additionally, other phenolic and flavonoid compounds were identified at lower concentrations, including catechin (143.5 µg/ g), coumaric acid (63.8 µg/ g), kaempferol (54.9 µg/ g), and hesperetin (43.4 µg/ g). The presence of these bioactive compounds together may lead to synergistic antimicrobial effects, improving the overall effectiveness of the extract against the phytopathogenic microorganisms.

Gas chromatography-mass spectrometry (GC-MS) analysis of the extract's secondary metabolites

Figure 4 and Table 2 summarize the results obtained from the GC-MS analysis. The collected data validated the existence of seven primary compounds, each was characterized by a distinct chemical structure and retention time. Hexadecanoic acid emerged as the predominant compound (26.4 %), observed at a retention time of 26.36 min. This fatty acid exhibits antimicrobial properties and may significantly contribute to the bioactivities observed in the extract. Phorbol was identified as the second most prevalent compound (16.4 %). Phorbol and its derivatives exhibit a range of biological activities, including antimicrobial effects, which may enhance the efficacy of the tested extract against the phytopathogens.

Table 1: *Analysis of phenolic and flavonoid compounds of methanolic extract of E. verticillata using high-performance liquid chromatography (HPLC).*

Compounds	Area	Concentration (µg/ g)
Gallic acid	430.61	2620.3
Caffeic acid	110.45	606.5
Syringic acid	123.61	580.9
Ferulic acid	61.53	265.3
Ellagic acid	15.97	202.6
Naringenin	27.29	198.4
Vanillin	60.89	177.8
Cinnamic acid	139.25	174.1
Quercetin	18.72	169.4
Catechin	8.64	143.5
Daidzein	33.41	136.8
Coumaric acid	29.97	63.8
Methyl gallate	16.97	63.3
Kaempferol	11.84	54.9
Hesperetin	12.07	43.4
Chlorogenic acid	Not detected	Not detected
Pyro catechol	Not detected	Not detected
Rutin	Not detected	Not detected
Apigenin	Not detected	Not detected

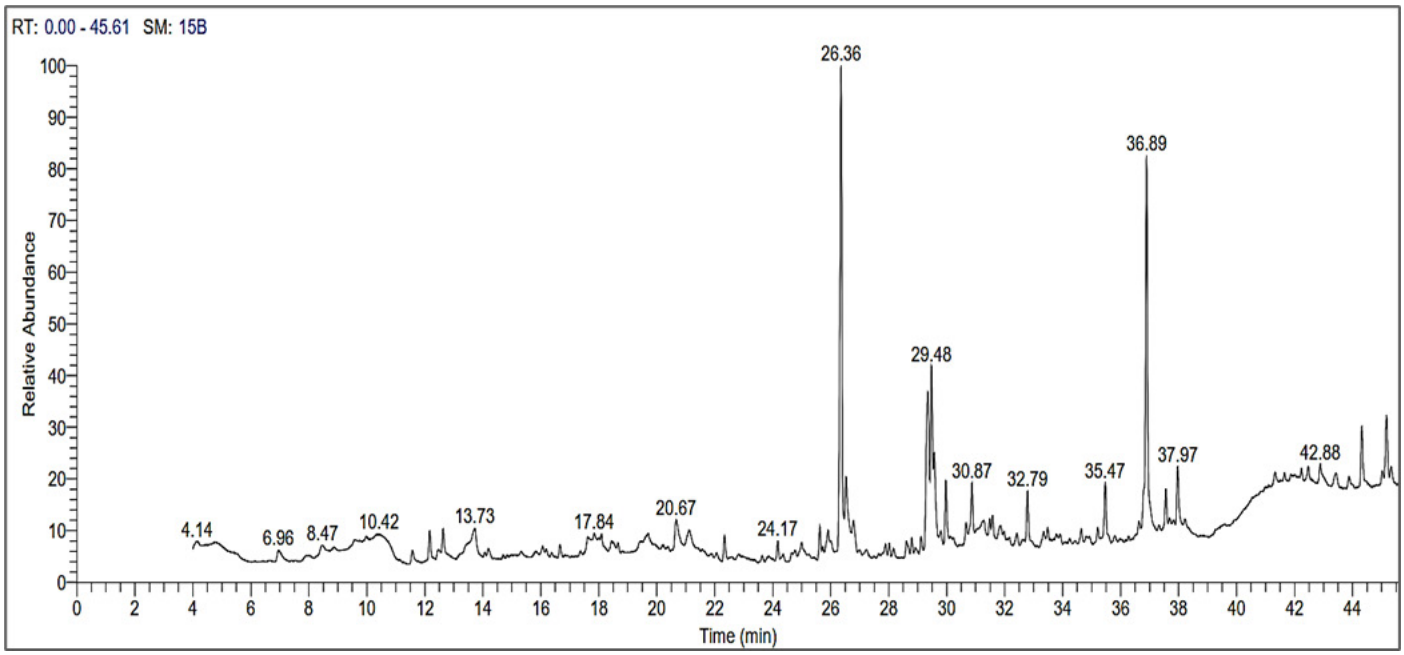


Figure 4: *Gas chromatography-mass spectrometry chromatogram of methanolic extract of Erica verticillata, showing the major detected bioactive compounds.*

Table 2: *List of chemical composition of the highest detected compounds in the methanol extract of E. verticillata leaves using gas chromatography–mass spectrometry (GC-MS) analysis.*

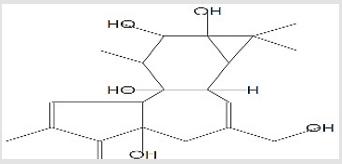
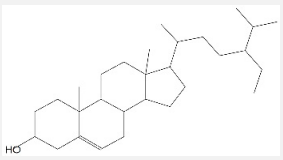
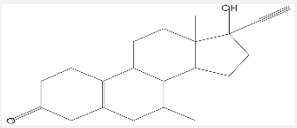
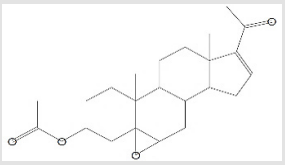
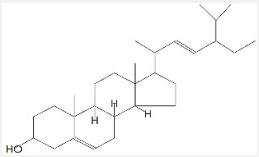
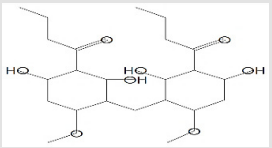
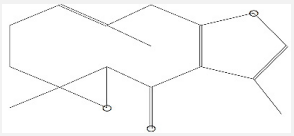
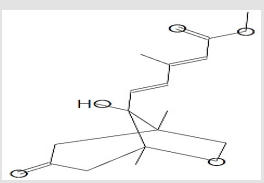
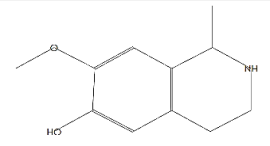
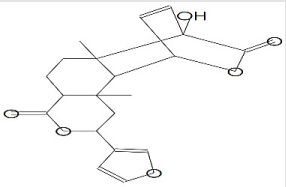
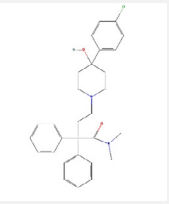
Compound	Retention time	Area %	Molecular formula	Molecular weight	PubChem compound identifier	Chemical Structure
Hexadecanoic acid	26.36	23.12	$C_{16}H_{32}O_2$	256	985	
Phorbol	36.89	16.41	$C_{20}H_{28}O_6$	364	442070	
trans-13-Octadecenoic acid	29.48	8.83	$C_{18}H_{34}O_2$	282	6161490	
9,12,15-Octadecatrienoic acid, (Z,Z,Z)-	29.36	8.19	$C_{18}H_{30}O_2$	278	5280934	
á-Sitosterol	45.17	3.30	$C_{29}H_{50}O$	414	222284	
Octadecanoic acid	29.97	2.95	$C_{18}H_{36}O_2$	284	5281	
Tibolone	35.47	2.89	$C_{21}H_{28}O_2$	312	444008	
Pregn-16-en-20-one,3-(acetyloxy)-5,6-epoxy-, (3á,5à,6à)-	37.97	2.88	$C_{23}H_{32}O_4$	372	22213077	
Stigmasterol	44.32	2.87	$C_{29}H_{48}O$	412	5280794	
Methylenebisdesaspidinol	30.87	2.48	$C_{23}H_{28}O_8$	432	613973	
Zederone	26.53	2.39	$C_{15}H_{18}O_3$	246	134687472	
Methyl phaseate	32.79	2.31	$C_{16}H_{22}O_5$	294	5363773	

Table continues on next page.....

Compound	Retention time	Area %	Molecular formula	Molecular weight	PubChem compound identifier	Chemical Structure
Salsoline	20.67	2.25	$C_{11}H_{15}NO_2$	193	46695	
Columbin	37.56	1.80	$C_{20}H_{22}O_6$	358	226036	
Loperamide	42.88	1.71	$C_{29}H_{33}ClN_2O_2$	476	3955	

Additional compounds were identified at moderate concentrations, comprising α-sitosterol, octadecenoic acid, tibolone, stigmasterol, and methyl phaseate. Figure 4 illustrates the relative area percentages of these compounds, offering a visual representation of their proportions in the extract. Other compounds, such as columbin and loperamide, were detected at lower concentrations. The variety of secondary metabolites in the *E. verticillata* extract suggests a complex phytochemical profile, contributing to its extensive antimicrobial effectiveness observed against several bacterial and fungal phytopathogens. The synergistic effect of these compounds probably enhances the extract's efficacy as a natural antimicrobial agent.

Discussion

Plant pathogens, including bacteria and fungi, are known to cause significant qualitative and quantitative harm to the crop production (Al-Askar *et al.*, 2025). Using plant extracts as alternative antimicrobial medicines has recently attracted considerable attention due to their efficacy, efficiency, and diminished side effects. The current study investigated the antimicrobial properties and chemical composition of the methanolic extract of *E. verticillata* concerning various phytopathogenic bacteria and fungi. The results indicated notable antibacterial and antifungal properties, as well as a range of bioactive compounds contributing to these observed effects. The results of the antibacterial assay suggested that the extract exhibited differing inhibitory effects against the examined phytopathogenic bacterial strains. The differential sensitivity levels observed among the bacterial strains indicate the specificity of the modes of action of the extract's bioactive compounds. This observed specificity may be attributed to variations in the cell wall composition and membrane permeability of the tested bacteria, affecting their susceptibility to the antimicrobial agents (Nazzaro *et al.*, 2013). A rrecent study demonstrated that plant extracts can serve as alternatives or adjuncts to conventional antibiotics, as their various compounds possess antimicrobial properties that may help combat antimicrobial resistance, a primary global health concern (Hernández-Bolaños *et al.*, 2025).

The antibacterial assay showed that *P. atrosepticum* and *S. scabiei* exhibited high sensitivity to the extract, even at a lowe concentration (300 µg/ mL), which holds considerable importance. *P. atrosepticum* is responsible for blackleg disease in potatoes, resulting in significant economic losses in global potato production (Czajkowski *et al.*, 2015). Similarly, *S. scabiei* is responsible for common scab disease in potato and other root crops, leading to diminished marketability and yield (Zhang *et al.*, 2024). The pronounced sensitivity of these pathogens to *E. verticillata* extract indicates promising avenues for developing natural bactericides to manage these economically significant plant diseases. The observation that certain bacterial strains, specifically *R. solanacearum* and *S. scabiei*, which exhibit resistance to conventional antibiotics, demonstrated notable sensitivity to the plant

extract at elevated concentrations (1000 µg/ mL) is particularly intriguing. This finding is consistent with recent studies that emphasized the potential of plant-derived compounds in addressing antibiotic-resistant bacteria (Angelini, 2024). The rise of antimicrobial resistance poses a growing challenge to human health and agriculture, making it particularly crucial to develop alternative antimicrobial agents (Salam *et al*., 2023; Ferraz, 2024). Our findings indicated that *E. verticillata* extract may represent a significant source of innovative antimicrobial compounds for tackling this issue. The absence of notable variations in inhibition zone diameters across the various extract concentrations suggests a saturation effect, implying that increasing the concentration beyond a specific limit does not improve the antibacterial activity (Al-Askar *et al*., 2025; Gaber *et al*., 2025).

In this study, the methanol extract exhibited significant antifungal efficacy against *B. cinerea*. This finding holds particular importance as *B. cinerea* is a necrotrophic fungal pathogen affecting many hosts, leading to gray mold disease in more than 200 plant species and causing considerable pre- and post-harvest losses (Hamdy *et al*., 2024). The significant antifungal activity demonstrated against *R. solani* and *F. oxysporum* highlights the extensive range of the antifungal activities of the extract. *R. solani* is a soil-borne pathogen responsible for various diseases, including damping-off, root rot, and stem canker, which affects many crops (Abo-Zaid *et al*., 2024). *F. oxysporum* is a complex of pathogenic strain responsible for vascular wilt diseases affecting various economically significant plants (Shehzadi *et al*., 2025). Successful suppression of these pathogens by *E. verticillata* extract indicates promising possibilities for creating natural fungicides with comprehensive disease management approaches (Salem *et al*., 2025). The exceptional antifungal efficacy of the plant extracts compared to copper hydroxide, is especially significant (Lamichhane *et al*., 2018). Copper-based fungicides have been extensively utilized in agriculture for many years; however, worries about their environmental persistence, soil accumulation, and adverse effects on non-target organisms have led to the exploration of alternative solutions (Lamichhane *et al*., 2018). The findings of this study suggest that *E. verticillata* extract may serve as an eco-friendly alternative to traditional copper-based fungicides, thereby promoting more sustainable agricultural practices. The mechanisms by which the extract exhibits antifungal properties may include the disruption of ergosterol; a crucial component of fungal cell membranes, or the inhibition of essential enzymes involved in the biosynthesis of fungal cell walls (Eliaš, Tóth Hervay, and Gbelská, 2024; Song *et al*., 2025).. The evidence suggests that numerous compounds derived from plants exhibit antifungal effects through similar mechanisms (Song *et al*., 2025; Xie *et al*., 2025). This finding is consistent with a recent study that showed methanolic extracts from various plant sources frequently exhibited potent antifungal effects by disrupting fungal cell wall synthesis or altering cellular metabolism, thereby inhibiting growth and development (Xie *et al*., 2025). To elucidate these effects, the phytochemical composition of the extract was analyzed.

High-performance liquid chromatography (HPLC) analysis revealed the presence of various phenolic and flavonoid compounds in the extract. The most abundant phenolic compounds identified were gallic acid (2620.3 µg/ g), caffeic acid (606.5 µg/g), and syringic acid (580.9 µg/ g). Additionally, naringenin (198.4 µg/ g) and quercetin (169.4 µg/ g) were noted as the predominant flavonoids. These results provide valuable insights into the potential bioactive compounds that contribute to the extract's antimicrobial activity. Gallic acid has been documented to exhibit potent antimicrobial properties against various bacterial and fungal pathogens (Flores-Maldonado *et al*., 2024). The mechanism of action involved disrupting the cell membrane, inhibiting vital enzymes, and chelating metal ions essential for microbial growth (Nasaj *et al*., 2024; Khwaza and Aderibigbe, 2025). The elevated levels of gallic acid in the extract probably play a crucial role in enhancing its antimicrobial effectiveness. Caffeic acid and syringic acid have shown antimicrobial properties in earlier studies (Ecevit *et al*., 2022; Beulah *et al*., 2025). Caffeic acid exhibits the ability to inhibit bacterial growth by disrupting cell membrane integrity and inhibiting essential cellular processes (Beulah *et al*., 2025). Syringic acid exhibits antimicrobial properties through comparable mechanisms and has been shown to enhance the efficacy of the standard antibiotics when used in conjunction (Ecevit *et al*., 2022). Naringenin and quercetin are flavonoids that exhibit well-documented antimicrobial properties (Shamsudin *et al*., 2022; Veiko *et al*., 2023; Hasnat *et al*., 2024). Naringenin can inhibit the growth of bacteria and fungi by disrupting cell membrane function and inhibiting

essential enzymes (Liu *et al.*, 2025). Quercetin exhibits antimicrobial activities through various mechanisms, including inhibition of DNA gyrase, disruption of cell membrane integrity, and interference with energy metabolism (Liang *et al.*, 2021). The presence of these flavonoids in the extract probably plays a significant role in its wide-ranging antimicrobial activity. A recent study on *E. manipuliflora* suggest that the genus *Erica* is rich in vanillic acid, fumaric acid, catechin hydrate, quercetin, and phloridzin dihydrate, all contributing to its antimicrobial potential (Yüksel *et al.*, 2021). This study revealed that *E. manipuliflora* extract exhibited effectiveness against several pathogenic microorganisms, such as *Staphylococcus aureus*, *Escherichia coli*, and *Salmonella typhimurium*, reinforcing the antimicrobial potential of the *Erica* genus.

The GC-MS analysis identified about 15 primary compounds in the extract, with hexadecanoic acid and phorbol being the most prevalent metabolites. This study also detected α-sitosterol, octadecenoic acid, tibolone, stigmasterol acid, and methyl phaseate as less prevalent metabolites. Hexadecanoic acid, also known as palmitic acid, is a saturated fatty acid recognized for its antimicrobial potential (Ganesan *et al.*, 2024). The mechanism of action involved disrupting the cell membrane, resulting in increased permeability and ultimately leading to cell death (Nasaj *et al.*, 2024; El-Bilawy *et al.*, 2025; Khwaza and Aderibigbe, 2025). Numerous studies detailed the direct beneficial effects of fatty acids, such as linoleic acid, palmitic acid, and their derivatives in managing phytopathogens (Zheng *et al.*, 2005; Liu *et al.*, 2008; Sumayo *et al.*, 2014; López-Arellanes *et al.*, 2025). Phorbol and its derivatives exhibit a range of biological activities, notably antimicrobial effects (Goel *et al.*, 2007). The high concentrations of these compounds in the extract may play a significant role in its antimicrobial efficacy. The variety of bioactive compounds found in the *E. verticillata* extract suggests that its antimicrobial activity is likely attributed to the combined effects of several compounds rather than relying on a single active principle (Guimarães and Venâncio, 2022; Vaou *et al.*, 2022; Gadouche *et al.*, 2023; Jabal *et al.*, 2025). This synergism may enhance overall efficacy and reduces the likelihood of resistance development, making plant extracts crucial in sustainable disease management strategies (Ayaz *et al.*, 2019). Before synthetic pesticides were introduced, the botanical pesticides were widely used in subsistence and commercial agriculture for a long time. The active components in botanical pesticides are naturally occurring substances extracted from plants using organic solvents and/or combined with essential pesticide adjuvants (Ngegba *et al.*, 2022). The effectiveness and efficacy of synthetic pesticides in managing crop diseases led to a gradual decline in the use of plant-based pesticide products. However, concerns about the environmental and health risks of the synthetic pesticides to humans have since emerged. (Chowdhury *et al.*, 2024; Shekhar *et al.*, 2024).

Finally, the findings of this study have important implications for sustainable plant disease management. The current antimicrobial efficacy of *E. verticillata* extracts against significant phytopathogens, and their enhanced performance relative to traditional antimicrobial agents in certain instances, highlights their potential as natural alternatives to the synthetic pesticides. Identifying specific bioactive compounds in the *E. verticillata* extract establishes a basis for subsequent studies on isolating and characterizing these compounds for potential commercial development as biopesticides. Moreover, comprehending the chemical composition of the extract may enhance the standardization and quality control of the botanical formulations, thereby addressing a significant challenge in the commercialization of plant-derived antimicrobial products.

Conclusions and Recommendations

This study demonstrated that methanolic extract of *Erica verticillata* leaves possess broad-spectrum antimicrobial activity against economically significant phytopathogens, including antibiotic-resistant bacterial strains (e.g., *Ralstonia solanacearum*) and fungi (*Botrytis cinerea*, *Fusarium oxysporum*). Notably, the extract outperformed the conventional copper hydroxide fungicides in inhibiting *B. cinerea*, recotrding 78.2 % vs. 58.7 %, respectively at 5000 µg/mL, highlighting its potential as a sustainable alternative to the synthetic agrochemicals. Phytochemical profiling revealed high concentrations of multiple bioactive compounds, including gallic acid (2620.3 µg/ g), hexadecanoic acid (26.4 %), and phorbol (16.4 %), which likely drive these amntimicrobial impacts through synergistic mechanisms, mainly membrane disruption and enzyme inhibition. Based on the strong antimicrobial efficacy demonstrated, it is recommended that *E. verticillata* methanolic extract

be further explored and developed as a natural biopesticide for integrated plant disease management. Its superior activity against key phytopathogens, especially *Botrytis cinerea*, positions it as a sustainable alternative to conventional agrochemicals.

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Novelty Statement

This study examined the bioactive potential of *Erica verticillata* methanol extract focusing on its phytochemical profile, and assessing its antibacterial and antifungal properties against various phytopathogenic microorganisms. The study provided a comprehensive evaluation of the bioactive compounds present in *E. verticillata* extract, highlighting their unique antimicrobial efficacy against phytopathogenic organisms for the first time. Moreover, it highlights the biocontrol prospects of the detected bioactive compounds and lays the groundwork for future applications as a natural source of antimicrobial agents for the sustainable management of plant diseases.

Author's Contribution

Abdallah Khalil, Karrar A. Hamzah, and Shimaa Bashir: Methodology, Investigations, and Writing the original draft. Said Behiry and Ahmed Abdelkhalek**:** Supervision, Statistical study design and analysis, Molecular data analysis, Revision and editing of the manuscript. All authors approved the final version of the manuscript.

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