Plant Protection (Scientific Journal of Agriculture)

49(1), Spring, 2026

10.22055/ppr.2026.50916.1855

Short Communication

Comparative antimicrobial efficacies and phytochemical profiling of three plant extracts against *Pseudomonas syringae*: a Preliminary study

M. Nair [1], D. Sayantan [*1]

1. ***Corresponding Author:** Department of Life Sciences, Christ University, Bengaluru – 560029, Karnataka, India (sayantan.d@christuniversity.in)

Received: 20 October 2025 Revised: 10 February 2026 Accepted: 13 February 2026

Abstract

This study evaluated the phytochemical profile and antimicrobial effects of extracts from *Colocasia esculenta*, *Ocimum tenuiflorum*, and *Murraya koenigii*. Well diffusion tests and minimum inhibitory concentration (MIC) testing revealed that the extracts of *Murraya koenigii* and *Ocimum tenuiflorum* exhibited comparable antibacterial activity against *Pseudomonas syringae*. Although this difference was not statistically significant, *Murraya* showed a numerically larger mean zone of inhibition (21.0±4.6 mm at 100 mg/mL) than *Ocimum* (13.3±1.2 mm). The least active was *Colocasia esculenta* with a zone of inhibition of 12.3±0.6 mm. Phytochemical screening revealed that all extracts included alkaloids, phenolics, flavonoids, and terpenoids. Although the experiment was qualitative, the GC-MS data clearly showed the main factors impacting antibacterial activity. While *Ocimum tenuiflorum* included significant phenolic and terpenoid compounds, including eugenol, caryophyllene, and linalool, *Murraya koenigii* contained carbazole alkaloids, such as mahanimbine and koenimbine. In contrast, *Colocasia esculenta* primarily include sterols and fatty acids, which are consistent with its lower inhibition zones and generally have less antibacterial qualities. A limitation of this study is that the active constituents were not isolated or quantitatively evaluated. These compounds from plants have potential as long term substitutes for synthetic antimicrobials, complementing sustainable agricultural methods. Further research is required to identify and characterise the active components for usage in agriculture and medicine. Thus, this study highlights the use of traditional medicinal plants to address the issues of existing antibiotic resistance and provide sustainable solutions.

Keywords: Plant-derived antimicrobials, well diffusion, bioactive substances, GC– MS analysis, sustainable agriculture

Associate editor: R. Rezaei (Ph.D.)

Citation: Nair, M., & Sayantan, D. (2026). Comparative antimicrobial efficacies and phytochemical profiling of three plant extracts against *Pseudomonas syringae*: a Preliminary study. *Plant Protection (Scientific Journal of Agriculture), 49*(1), 117-126. https://doi.org/10.22055/ppr.2026.50916.1855

گیاه‌پزشکی (مجله علمی کشاورزی)
جلد ۴۹، شماره ۱، بهار ۱۴۰۵

10.22055/ppr.2026.50916.1855

گزارش کوتاه انگلیسی

بررسی اثرات ضدمیکروبی و مشخصات فیتوشیمیایی سه عصاره گیاهی بر باکتری *Pseudomonas syringae*: مطالعه‌ای مقدماتی

مگانا نایر[1]، دی. سایانتان [1*]

۱- * **نویسنده مسوول:** دانشکده علوم زیستی، دانشگاه کرایست، بنگلور – ۵۶۰۰۲۹، کارناتاکا، هند

تاریخ دریافت: ۱۴۰۴/۰۷/۲۸ تاریخ بازنگری: ۱۴۰۴/۱۱/۲۱ تاریخ پذیرش: ۱۴۰۴/۱۱/۲۴

چکیده

مطالعه حاضر با هدف ارزیابی مشخصات فیتوشیمیایی و پتانسیل ضدمیکروبی عصاره‌های گیاهی *Colocasia esculenta*، *Ocimum tenuiflorum* و *Murraya koenigii* انجام شد. نتایج حاصل از آزمون انتشار در چاهک و تعیین حداقل غلظت مهارکنندگی (MIC) نشان داد که عصاره‌های *Murraya koenigii* و *Ocimum tenuiflorum* فعالیت ضدباکتریایی قابل‌توجهی را علیه *Pseudomonas syringae* بروز می‌دهند. هرچند تفاوت مشاهده‌شده از نظر آماری معنی‌دار نبود، اما میانگین قطر هاله عدم رشد برای *Murraya* (۴/۶ ± ۲۱ میلی‌متر در غلظت ۱۰۰ میلی‌گرم بر میلی‌لیتر) نسبت به *Ocimum* (۱/۲ ± ۱۳/۳ میلی‌متر) برتری عددی نشان داد. کمترین اثر مهاری مربوط به *Colocasia esculenta* با میانگین قطر ۰/۶±۱۲/۳ میلی‌متر بود. غربالگری‌های فیتوشیمیایی، حضور آلکالوئیدها، ترکیبات فنولی، فلاونوئیدها و ترپنوئیدها را در تمامی عصاره‌ها تأیید نمود. اگرچه ماهیت ارزیابی آزمایش در این بخش کیفی بود، اما داده‌های حاصل از آنالیز GC-MS، مؤلفه‌های کلیدی دخیل در فعالیت ضدباکتریایی را به‌روشنی تفکیک کرد؛ به‌طوری که *Ocimum tenuiflorum* سرشار از ترکیبات فنولی و ترپنوئیدی نظیر Eugenol، Caryophyllene و Linalool بود، در حالی که در *Murraya koenigii* آلکالوئیدهای carbazole از قبیل Mahanimbine و Koenimbine شناسایی گردید. در مقابل، مشخصات شیمیایی *Colocasia esculenta* عمدتاً متشکل از استرول‌ها و اسیدهای چرب بود که با فعالیت مهاری کمتر این گیاه همخوانی دارد و به‌طور کلی خاصیت ضدباکتریایی ضعیف‌تری را نشان می‌دهد. از محدودیت‌های این پژوهش، عدم جداسازی و ارزیابی کمی اجزای فعال می‌باشد. ترکیبات زیست‌فعال گیاهی به دلیل پتانسیل بالا، جایگزین‌های مناسبی برای عوامل ضدمیکروبی مصنوعی در راستای تحقق اهداف کشاورزی پایدار محسوب می‌شوند. انجام مطالعات تکمیلی جهت شناسایی و تعیین مشخصات دقیق ترکیبات فعال برای کاربردهای بالینی و کشاورزی ضروری است. در نهایت، این مطالعه بر کاربرد گیاهان دارویی سنتی به عنوان راهکاری نوین جهت مقابله با معضل مقاومت‌های آنتی‌بیوتیکی و دستیابی به سیستم‌های پایدار تأکید دارد.

کلید واژه‌ها: ضدمیکروب‌های گیاهی، انتشار در چاهک، ترکیبات زیست‌فعال، آنالیز GC-MS، کشاورزی پایدار

دبیر تخصصی: دکتر رسول رضائی

Introduction

The emergence of antibiotic resistance in plant pathogens is one of the main challenges posed to sustainable agriculture. The Gram-negative bacterium, *Pseudomonas syringae* causes harmful diseases, such as leaf spots, blossom blast, and stem cankers in fruit trees, especially in cool, humid environments (Kennelly et al., 2007). Its pathogenicity is enhanced by environmental variables that promote apoplastic hydration and type III secretion effectors (Xin et al., 2018). Conventional management makes use of antibiotics, such as oxytetracycline and streptomycin; however, resistance continues to rise, and chemical residues contaminate ecosystems and endanger human health (Córdova et al., 2023).

Plant-based antimicrobials offer a sustainable alternative, due to their low toxicity, biodegradability, and multi-target mechanisms, which can delay the emergence of resistance. The International Goals for Ecological Farming state that bioactive compounds with antibacterial properties, such as terpenoids and alkaloids, are found in medicinal plants, including *Murraya koenigii* and *Ocimum tenuiflorum* (Alattas et al., 2024). Despite this potential, few research studies have evaluated their efficacy against *P. syringae* using standardized methods, such as phytochemical profiling and minimum inhibitory concentration (MIC) assays (Soltaniband et al., 2024).

Despite research showing antibacterial action in plants such as *Quercus robur* and *Tsuga canadensis*, *O. tenuiflorum*, *M. koenigii*, and *Colocasia esculenta* have not been directly compared to *P. syringae*. Specifically, no work has integrated well – diffusion testing, MIC determination, and GC-MS-based phytochemical analysis to identify the optimal botanical extract and its bioactive components for biocontrol applications.

This study intends to bridge the gap by carefully assessing the antibacterial activity of ethanolic extracts from *M. koenigii*, *O. tenuiflorum*, and *C. esculenta* against that of *P. syringae*. We used well – diffusion experiments, MIC tests, and GC-MS analysis to assess the efficacy and phytochemical profiles of potential plant-based, sustainable pesticide substitutes. The findings support integrated pest management strategies in agriculture to aid the development of effective biopesticides.

Materials and Methods

Plant Collection and Authentication

We obtained fresh *Ocimum tenuiflorum* (holy basil) leaves from Bengaluru, Karnataka, as well as *Murraya koenigii* (curry leaves) and *Colocasia esculenta* (taro leaves) from local markets in Ahmedabad, Gujarat. The samples were transported at 4°C for verification by a certified botanist from the Central Ayurveda Research Institute (CARI), Bengaluru. The leaves were cleaned, shade-dried at 25 to 30°C for seven to ten days, ground into a fine powder (less than 0.4 mm), and stored at 4°C in airtight containers.

Bacterial Strain, Culture Conditions and Inoculum Preparation

A lyophilized culture of *Pseudomonas syringae* (MTCC 1604) from the Microbial Type Culture Collection (MTCC), Chandigarh, was revived in nutrient broth and incubated at 28 °C for 24 to 48 hours. The purity of the culture was confirmed using UV-induced fluorescence and Gram staining. Ethanolic extracts of plant samples were obtained using Soxhlet extraction, which involved extracting 10 g of dried plant powder from each sample, with 200 mL of 70% ethanol until the solvent turned colorless. Concentration of the extracts was achieved by rotary evaporation at 40 °C and 100 rpm. 100 mg/mL stock solutions were prepared in 70% ethanol and stored at 4 °C until needed (Faradilla et al., 2024). For the inoculum, a single colony from a nutrient

agar plate was inoculated in 25 mL of nutrient broth for 18 to 24 hours at 120 rpm and 28 °C. After that, sterile nutritional broth was used to dilute the bacterial suspension to a 0.5 McFarland standard (≈ 1.5 x 10^2 CFU/mL) (Popović et al., 2021).

Antimicrobial Activity Assay

Antibacterial activity was determined using the well diffusion method. A standardized *P. syringae* suspension (100 µL) was used to swab Mueller-Hinton agar plates. A sterile cork borer was used to create wells with an 8 mm diameter. Concentrations of 25, 50, 75, and 100 mg/mL of plant extract were tested. The positive and negative controls were 70% ethanol and streptomycin (1000 µg/mL). After an hour of pre-diffusion at 4°C, the plates were incubated at 28°C for 24 hours. Zones of inhibition were measured in triplicate.

Determination of Minimum Inhibitory Concentration (MIC)

Broth microdilution in 96-well plates was used to calculate the MIC (Vanegas et al., 2021). Each extract was serially diluted twice, using Mueller-Hinton broth, ranging from 100 mg/mL to 0.195 mg/mL. A standardized bacterial solution (100 µL) was added to each well. Growth control (broth with bacteria), sterility control (broth alone), and solvent control (ethanol with bacteria) were among the controls. Plates were evaluated for bacterial growth, using both visual examination and OD measurement after an incubation period of 18 to 24 hours at 37°C. The minimal inhibitory concentration (MIC) was defined as the concentration at which no significant growth occurred.

Phytochemical Screening

The main kinds of bioactive compounds found in the plant extracts were identified using standard methods in qualitative phytochemical analysis. To detect alkaloids, Wagner's reagent was applied to 0.5 mL of each extract; the formation of a reddish-brown precipitate indicated the presence of alkaloids. Phenols and tannins were identified using the ferric chloride test, which caused a bluish-black color shift when $FeCl_3$ was added to the extract. The presence of flavonoids was indicated by the extract's color changing from yellow to colorless upon the addition of 10% NaOH and diluted HCl.

Saponins were identified using the foam test. It involved creating persistent foam by quickly shaking 0.5 mL of extract with 2 mL of distilled water. Steroids were detected through the Liebermann-Burchard test, which involved mixing 0.5 mL of extract with concentrated H_2SO_4 and chloroform. Steroids were detected by a shift in color from red to blue-green. Quinones were detected by a blue-green or red color shift when 1% NaOH was added to the extract. Carbohydrates were detected using Benedict's test, which produced a reddish-brown precipitate when the extract was heated with Benedict's reagent (Ramya et al., 2015; Dubale et al., 2023). All reactions were performed in triplicate, and results were recorded based on color intensity and precipitate formation, with scoring indicating weak (+), moderate (++), or strong (+++).

GC–MS Analysis

Plant samples were phytochemically profiled at Aknowlytiq Academy in Bengaluru, India, using a GCMS-QP2010 SE system (Shimadzu, Japan) and gas chromatography–mass spectrometry (GC–MS) analysis. A 1 µL sample volume was injected for analysis. Substances were identified by comparing the obtained mass spectra with standard volatile chemicals and the laboratory's internal reference libraries. The chromatographic settings, data acquisition, and separation parameters were operated in accordance with the analytical facility's regular operating procedures.

Statistical Analysis

The data was analyzed using a mixed-model ANOVA in SPSS v21, with extract

type and concentration working as fixed factors and replication working as a random component. For post-hoc comparisons, Tukey's and Bonferroni tests were used ($\alpha = 0.05$).

Results and Discussion

The relative antibacterial efficacy of ethanolic extracts from *Murraya koenigii*, *Ocimum tenuiflorum*, and *Colocasia esculenta* against the phytopathogen *Pseudomonas syringae* was carefully assessed using integrated microbiological and phytochemical techniques. This study used the well – diffusion assay to identify inhibitory zones across a concentration gradient, the broth microdilution method to ascertain the minimum inhibitory concentration (MIC), and a range of biochemical and chromatographic techniques to understand the underlying phytochemical profiles. The results demonstrate a distinct hierarchy of antibacterial activity that is strongly associated with the distinct secondary metabolite composition of each plant extract.

Significant differences in activity among the three plant extracts were found by analyzing the well-diffusion assay data. With a statistically significant interaction term (ANOVA, $p<0.001$), Table 1 shows that the antibacterial efficacy was dependent on both extract and concentration. *Murraya koenigii* extract produced the largest mean zone of inhibition (21.0±4.6 mm) at the highest tested concentration of 100 mg/mL. *Ocimum tenuiflorum* (13.3±1.2 mm) and *Colocasia esculenta* (12.3±0.6 mm) came next. While the negative control (70% ethanol) had no effect, the positive control (streptomycin) had the greatest inhibition (25.1±0.8 mm), indicating the bacterial strain's susceptibility.

Each extract revealed a dose-response relationship, with inhibitory zones growing with higher doses ($p<0.01$). A non-monotonic response was observed for *Ocimum tenuiflorum* and *Colocasia esculenta*, with inhibition zones decreasing at higher concentrations. This is likely to reflect methodological factors rather than true loss of activity. Volatile phenolics in *O. tenuiflorum* may partially evaporate, and higher extract loads can increase viscosity or aggregation, limiting diffusion. Similarly, the fatty-acid-rich *C. esculenta* extract may spread unevenly in agar. Minor variations in well size or agar thickness can also affect results, as reported in other plant extract diffusion studies.

Tukey's HSD test was used in post-hoc analysis to determine the statistical relationships between the extracts. Crucially, *Murraya koenigii* displayed a numerically larger mean inhibition zone than *Ocimum tenuiflorum*; however, this difference was not statistically significant ($p=0.156$), indicating that both species performed similarly well in the diffusion assay under these experimental conditions. Both *M. koenigii* and *O. tenuiflorum* formed a statistically superior group with better inhibition ($p<0.001$) than *C. esculenta*. At lower concentrations (25 and 50 mg/mL), the inhibitory zones produced by *O. tenuiflorum* and *C. esculenta* formed a homogenous subgroup, whereas *M. koenigii* showed a noticeably higher level of activity even at these lower dosages.

The broth microdilution method provided a more precise assessment of antibacterial effectiveness by determining the Minimum Inhibitory Concentration (MIC). The assay findings showed a significant and considerable correspondence with the diffusion data. At the same MIC value of 0.195 mg/mL, the extracts from *Ocimum tenuiflorum* and *Murraya koenigii* prevented observable *P. syringae* growth in each triplicate trial.

This result shows that both extracts had nearly identical intrinsic bacteriostatic activity against this pathogen despite differences in their chemical composition, quantitatively confirming the statistical

equality (p=0.156) observed in the well – diffusion assay. Despite its widespread use, the two-fold serial dilution technique restricts specificity; hence, even if the true MICs for both may be marginally below 0.195 mg/mL, they are equivalent within the limitations of the test. *M. koenigii* showed slightly more variability in optical density measurements at the MIC endpoint (OD600 = 0.078±0.056) than *O. tenuiflorum* (OD600 = 0.049±0.003).

This might indicate slight changes in the stability of the molecule or differences in the inhibitory kinetics. However, *Colocasia esculenta* extract did not completely inhibit at all of the tested dosages, which ranged from 100 mg/mL to 0.024 mg/mL. This implies that the extract has a fundamentally weaker relationship with the bacterial cells, where it may simply inhibit growth or require much higher doses or different conditions to have a noticeable impact. It also highlights the extract's poor efficacy as a stand-alone bactericide in comparison to the other two plants.

To understand the molecular bases of the claimed antibacterial action, a thorough phytochemical profile was conducted. The first qualitative screening revealed that *M. koenigii* tested very positive for phenolics/tannins, *O. tenuiflorum* for flavonoids and saponins, and *C. esculenta* for alkaloids, flavonoids, saponins, and steroids (Table 2). All three have noticeable levels of steroids. This preliminary research suggested the presence of a number of bioactive compounds.

Gas Chromatography-Mass Spectrometry (GC-MS) analysis provided conclusive, compound-specific information that accurately matched and explained the bioassay results. Figure 1 displays the combined total ion chromatogram (TIC). Carbazole alkaloids, a family of substances with proven bioactivity, were included in *Murraya Koenigii*'s profile. The relative abundance of 14.41% for mahanimbine was the most noteworthy finding. This is consistent with earlier research that identified *M. koenigii* as a rich source of these alkaloids (Balakrishnan et al., 2020). Carbazole alkaloids, such as mahanimbine, are known to have antibacterial properties by intercalating into and rupturing the integrity of bacterial cell membranes.

Table 1 Mean inhibition zones of plant extracts against *Pseudomonas syringae*.

Concentration	*Murraya koenigii* (mm)	*Ocimum tenuiflorum* (mm)	*Colocasia esculenta* (mm)	Streptomycin (mm)	70% ethanol (mm)
25 mg/mL	16.3±4.0^{b}	10.7±0.6^{ab}	7.0±5.6^{b}		
50 mg/mL	18.0±2.0^{b}	12.0±0.0^{a}	3.7±6.4^{b}		
75 mg/mL	17.7±2.3^{b}	9.0±7.0^{b}	4.0±6.9^{b}	25.1±0.8	0±0
100 mg/mL	21.0±4.6^{a}	13.3±1.2^{a}	12.3±0.6^{a}		

Values are mean ± SD (n = 3). Different superscript letters within the same column indicate statistically significant differences among concentrations (Tukey's post-hoc test, $p < 0.05$).

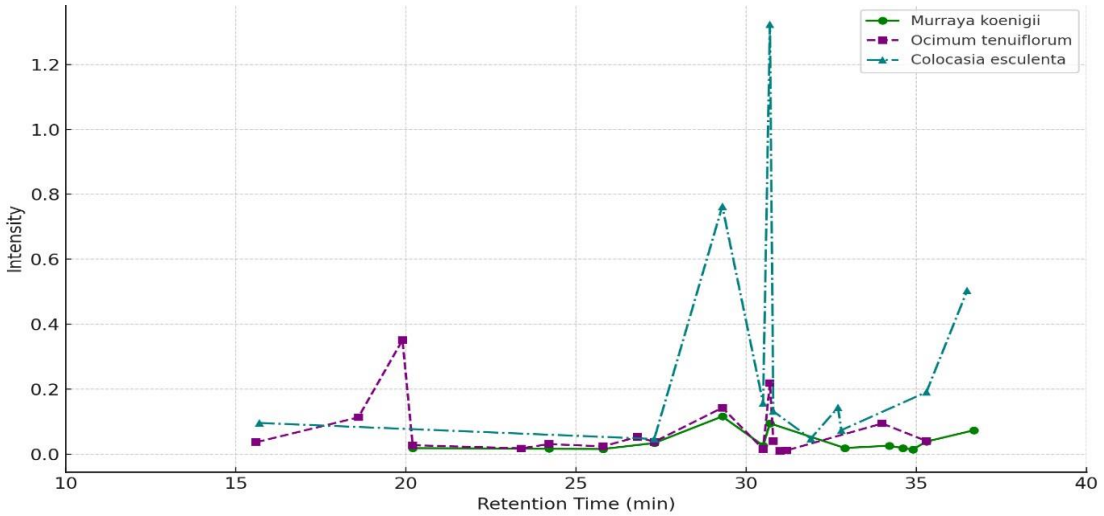


Figure 1. Combined TIC of ethanol extracts of *Murraya koenigii, Ocimum tenuiflorum*, and *Colocasia esculenta* by GC – MS, showing major bioactive compounds relevant to antimicrobial analysis.

This results in cell death by increasing permeability and allowing cellular contents to leak out (Viteritti et al., 2022). This technique is particularly effective against Gram-negative bacteria, such *P. syringae*, whose pathogenicity depends on their outer membrane acting as a substantial barrier (Collmer et al., 2000). The *M. koenigii* extract also contained significant levels of synergistic chemicals, including caryophyllene (3.54%) and n-Hexadecanoic acid (22.75%). By changing the fluidity of the membrane and allowing more bioactive molecules to flow through, these substances may increase activity (Rajendran et al., 2014).

This multi-target, synergistic action has several advantages, one of which is that it reduces the likelihood that infections will rapidly develop resistance, which is a significant issue with single-site synthetic pesticides.

Ocimum tenuiflorum's phytochemical profile was dominated by phenolic compounds, especially methyleugenol (27.81%) and eugenol (8.95%), which is consistent with its classification as an aromatic medicinal plant. Strong antibacterial qualities have been demonstrated for phenols. They induce cellular malfunction and death by denaturing vital proteins and enzymes, rupturing microbial cell walls and membranes, and altering proton motive forces (Chutimanukul et al., 2022).

The high concentration of these phenolics provides a biological explanation for the *O. tenuiflorum* extract's strong and consistent effectiveness against *P. syringae*. It demonstrates how two distinct phytochemical pathways, one using membrane-lytic alkaloids and the other using phenolics that disrupt enzymes - can work together to produce comparable antibacterial action against the same target organism. This convergence supports the idea of developing combinatorial botanical formulations, in which several mechanisms of action work together to further limit the emergence and spread of resistant disease strains (Varghese et al., 2018).

However, the GC-MS analysis of *Colocasia esculenta* revealed a profile dominated by fatty acids, which together formed a considerable share (61.5%) of the compounds detected, with linolenic acid (9,12,15-Octadecatrienoic acid) accounting for 38.07%. While some fatty acids are

known to have bacteriostatic properties, their immediate effects are typically less disruptive than the rapid membrane lysis caused by carbazole alkaloids or the potent enzyme inhibition by high-concentration phenolics. This is because fatty acids typically, gradually integrate into and alter the lipids present in microbial membranes. The conflicting results between the two studies can be explained by the large dose (100 mg/mL) used in the well-diffusion test, which was sufficient to generate a limited zone of growth suppression, most likely through a cumulative bacteriostatic action. However, in the broth microdilution MIC experiment, which measures a complete halt in growth over a liquid medium, the fatty acid-dominated extract failed to achieve a distinct tidal endpoint within the indicated range.

Furthermore, the physical properties of the *C. esculenta* extract, likely to be more viscous due to its fatty acid content - may have prevented its uniform diffusion in the agar matrix, leading to the smaller and more diverse inhibitory zones observed (Tosif et al., 2022).

This study has a number of limitations. Although useful for screening, the well-diffusion assay revealed significant variation in inhibitory zone diameters, especially for *O. tenuiflorum* at 75 mg/mL (SE = 7.0 mm) and *C. esculenta* at different dosages. The physicochemical characteristics of the extracts are the reason for this variation. Eugenol and methyleugenol, two significant phenolics in *O. tenuiflorum*, are volatile and may cause unequal evaporation and dispersion during plate preparation (Houdkova et al., 2020). Similarly, the fatty acid-rich *C. esculenta* extract's viscosity and poor water miscibility may prevent it from diffusing through agar, resulting in uneven zone edges and widths. Despite conventional methods of operation, these effects might have been increased by small technical differences in well structure and agar thickness.

These aspects show how diffusion-based techniques are inherently constrained by the physical characteristics of plant extracts (Holder & Boyce, 1994). Additionally, all the experiments were carried out in extremely controlled in – vitro settings. It is very challenging to translate these findings to field conditions, due to variables like UV deterioration, precipitation, temperature swings, and intricate soil-plant-microbe interactions. It remains necessary to assess these crude extracts' phytotoxicity, stability, and durability in field settings. The findings suggest that *M. koenigii* and *O. tenuiflorum* have great potential as long-term, plant-based alternatives to conventional copper-based or antibiotic treatments for managing bacterial diseases in agriculture (Pacyga et al., 2021).

Acknowledgments

The authors thank Christ University for its facilities, Aknowlytiq Academy for GC– MS analysis, MTCC for the *Pseudomonas syringae* culture, and CARI for plant authentication.

References

Alattas, H., Glick, B.R., Murphy, D.V., and Scott, C. (2024). Harnessing *Pseudomonas* spp. for sustainable plant crop protection, *Frontiers in microbiology*, *15*, 1485197. https://doi.org/10.3389/fmicb.2024.1485197

Balakrishnan, R., Vijayraja, D., Jo, S.– H., Ganesan, P., Su– Kim, I., and Choi, D.– K. (2020). Medicinal profile, phytochemistry, and pharmacological activities of *Murraya koenigii* and its primary bioactive compounds, *Antioxidants, 9*(2), 101. https://doi.org/10.3390/antiox9020101

Chutimanukul, P., Jindamol, H., Thongtip, A., Korinsak, S., Romyanon, K., Toojinda, T., Darwell, C. T., Wanichananan, P., Panya, A., Kaewsri, W., Auvuchanon, A., Mosaleeyanon, K., & Chutimanukul, P. (2022). Physiological responses and variation in secondary metabolite content among Thai holy basil cultivars (*Ocimum tenuiflorum* L.) grown under controlled environmental conditions in a plant factory. *Frontiers in Plant Science, 13*, 1008917. https://doi.org/10.3389/fpls.2022.1008917

Collmer, A., Badel, J. L., Charkowski, A. O., Deng, W.-L., Fouts, D. E., Ramos, A. R., Rehm, A. H., Anderson, D. M., Schneewind, O., van Dijk, K., & Alfano, J. R. (2000). *Pseudomonas syringae* Hrp type III secretion system and effector proteins. *Proceedings of the National Academy of Sciences, 97*(16), 8770–8777. https://doi.org/10.1073/pnas.97.16.8770

Córdova, P., Rivera– González, J.P., Rojas– Martínez, V., Fiore, N., Bastías, R., Zamorano, A., Vera, F., Barrueto, J., Díaz, B., Ilabaca– Díaz, C., Bertaccini, A., and Higuera, G. (2023). Phytopathogenic *Pseudomonas syringae* as a threat to agriculture: Perspectives of a promising biological control using bacteriophages and microorganisms, *Horticulturae*, *Horticulturae, 9*(6), 712. https://doi.org/10.3390/horticulturae9060712

Dubale, S., Kebebe, D., Zeynudin, A., Abdissa, N., and Suleman, S. (2023). Phytochemical screening and antimicrobial activity evaluation of selected medicinal plants in Ethiopia, *Journal of experimental pharmacology*, 51-62. https://doi.org/10.2147/JEP.S379805

Faradilla, M., Fidrianny, I., and Iwo, M.I. (2024). Antioxidant and immunomodulatory activities of ethanol extracts from *Syzygium cumini* L. Skeels and *Pogostemon cablin* Benth, *Narra J, 4*(3), e918. https://doi.org/10.52225/narra.v4i3.015

Holder, I. A., & Boyce, S. T. (1994). Agar well diffusion assay testing of bacterial susceptibility to various antimicrobials in concentrations non-toxic for human cells in culture. *Burns, 20*(5), 426–429. https://doi.org/10.1016/0305-4179(94)90035-3

Houdkova, M., Albarico, G., Doskocil, I., Tauchen, J., Urbanova, K., Tulin, E. E., & Kokoska, L. (2020). Vapors of volatile plant-derived products significantly affect the results of antimicrobial, antioxidative, and cytotoxicity microplate-based assays. *Molecules, 25*(24), 6004. https://doi.org/10.3390/molecules25246004

Kennelly, M. M., Cazorla, F. M., de Vicente, A., Ramos, C., & Sundin, G. W. (2007). *Pseudomonas syringae* diseases of fruit trees: Progress toward understanding and control. *Plant Disease, 91*(1), 4–17. https://doi.org/10.1094/PD-91-0004

Pacyga, K., Ronga, D., & Michalak, I. (2021). Plant extracts—importance in sustainable agriculture. *Italian Journal of Agronomy, 16*(2), 1851. https://doi.org/10.4081/ija.2021.1851

Popović, T., Menković, J., Prokić, A., Zlatković, N., and Obradović, A. (2021). Isolation and characterization of *Pseudomonas syringae* isolates affecting stone fruits and almond in

Montenegro, *Journal of Plant Diseases and Protection*, *128*(2), 391-405. https://doi.org/10.1007/s41348– 020– 00417– 8

Rajendran, M.P., Pallaiyan, B.B., and Selvaraj, N. (2014). Chemical composition, antibacterial and antioxidant profile of essential oil from *Murraya koenigii* (L.) leaves, *Avicenna journal of phytomedicine, 4*(3), 200.

Ramya, B., Malarvili, T., & Velavan, S. (2015). GC–MS analysis of bioactive compounds in *Bryonopsis laciniosa* fruit extract. *International Journal of Pharmaceutical Sciences and Research, 6*(8), 3375–3379. https://doi.org/10.13140/RG.2.2.20114.30401

Soltaniband, V., Barrada, A., Delisle– Houde, M., Dorais, M., Tweddell, R.J., and Michaud, D. (2024). Forest tree extracts induce resistance to *Pseudomonas syringae* pv. *tomato* in *Arabidopsis*, *Scientific Reports, 14*(1), 24726. https://doi.org/10.1038/s41598– 024– 74576– 1

Tosif, M. M., Najda, A., Klepacka, J., Bains, A., Chawla, P., Kumar, A., Sharma, M., Sridhar, K., Gautam, S. P., & Kaushik, R. (2022). A concise review on taro mucilage: Extraction techniques, chemical composition, characterization, applications, and health attributes. *Polymers, 14*(6), 1163. https://doi.org/10.3390/polym14061163

Vanegas, D., Abril– Novillo, A., Khachatryan, A., Jerves– Andrade, L., Peñaherrera, E., Cuzco, N., Wilches, I., Calle, J., and León– Tamariz, F. (2021). Validation of a method of broth microdilution for the determination of antibacterial activity of essential oils, *BMC Research Notes, 14*(1), 439. https://doi.org/10.1186/s13104– 021– 05838– 8.

Varghese, A., Mysore Babu, H., & Kukkera, P. N. (2018). Comparative evaluation of efficacy *of Murraya koenigii* and chlorhexidine gluconate in the treatment of gingivitis: A randomized controlled clinical trial. *Journal of Indian Society of Periodontology*, *22*(5), 427–432. https://doi.org/10.4103/jisp.jisp_112_18

Viteritti, E., Oliva, E., Eugelio, F., Fanti, F., Palmieri, S., Bafile, E., Compagnone, D., & Sergi, M. (2022). Analysis of carbazole alkaloids in *Murraya koenigii* by high-performance liquid chromatography coupled to tandem mass spectrometry with a predictive multi-experiment approach. *Journal of Chromatography Open, 2*, 100055. https://doi.org/10.1016/j.jcoa.2022.100055

Xin, X.– F., Kvitko, B., and He, S.Y., *Pseudomonas syringae*. (2018). What it takes to be a pathogen, *Nature Reviews Microbiology, 16*(5), 316-328. https://doi.org/10.1038/nrmicro.2018.17

© 2026 by the authors. Licensee SCU, Ahvaz, Iran. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution-NonCommercial 4.0 International (CC BY-NC 4.0 license) (http://creativecommons.org/licenses/by-nc/4.0/.