International Journal of Agricultural and Applied Sciences, June 2026, 7(1): 55-62
https://www.agetds.com/ijaas
ISSN: 2582-8053
https://doi.org/10.52804/ijaas2026.716

Research Article

OPEN ACCESS

In Vitro Antimicrobial Activities of Leaf Extracts of *Mangifera indica* Dehnh. and *Psidium guajava* F. Muell. Against Selected Antibiotic-Resistant Bacteria Strains

Julius Oluwatosin AYOADE[1*]; Oluwarotimi John JOSEPH[1]; Habeebat Eniola AROYEHUN[1]; Oluwadamiloju Mofeoluwa ADEKUNLE[1]; Abdullahi Adeyemi ADEGOKE[2]

[1]*Department of Microbiology, Faculty of Science, University of Ibadan, Ibadan, Nigeria*
[2]*Department of Pharmacognosy and Drug Development, Faculty of Pharmaceutical Science, Mercy Medical University, Iwara, Iwo, Osun State, Nigeria*
**Corresponding author e-mail: ayoadejulius8@gmail.com*
(Received: 05/03/2026; Revised: 19/05/2026; Accepted: 10/06/2026; Published: 20/06/2026)

ABSTRACT
The global rise of antibiotic-resistant bacteria necessitates the discovery of new antimicrobial agents. This study investigated the antibacterial potential of *Mangifera indica* and *Psidium guajava* leaf extracts against multidrug-resistant pathogens, including *Escherichia coli*, *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Pseudomonas aeruginosa*. Leaves were extracted using methanol, ethanol, and aqueous solvents. Phytochemical analysis confirmed the presence of alkaloids, flavonoids, saponins, and other bioactive compounds, with methanol extracts yielding the most comprehensive profile. Alkaloids were the most abundant phytochemical. Antimicrobial testing revealed that methanol extracts were the most effective, demonstrating the largest zones of inhibition against *S. aureus* and *P. aeruginosa* at a concentration of 100 g/mL. Ethanol extracts also showed significant activity, while aqueous extracts were moderately effective. Determination of the Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) confirmed that methanol extracts had the lowest inhibitory values, indicating superior bactericidal potential. Furthermore, Gram-positive bacteria (*S. aureus* and *S. epidermidis*) were more susceptible to the extracts than Gram-negative strains, likely due to differences in cell wall structure. The results highlight the efficacy of *M. indica* and *P. guajava* extracts, particularly methanol-based ones, against resistant bacteria. The study concludes that the bioactive compounds in these plants are promising candidates for development as alternative antimicrobial agents.
Keywords: *Mangifera indica*, *Psidium guajava*, Antibiotic resistance, Phytochemicals, Antibacterial activity

INTRODUCTION

Antimicrobial resistance (AMR) refers to the ability of microorganisms to survive and proliferate despite exposure to antimicrobial agents. This phenomenon develops through various genetic and biochemical mechanisms, including spontaneous mutation and horizontal gene transfer, enabling microorganisms to evade the action of antibiotics (Hughes and Andersson, 2017). The widespread misuse and overuse of antibiotics in human and veterinary medicine have accelerated the emergence of multidrug-resistant (MDR) pathogens, posing a serious challenge to global public health (Zilberberg et al., 2016). The growing dissemination of antimicrobial resistance genes has further complicated the treatment of infectious diseases and increased healthcare costs worldwide (Morar and Wright, 2010). Consequently, the development of alternative antimicrobial strategies has become an urgent priority (Ficker et al., 2003; Lewis and Ausubel, 2006).

Medicinal plants have historically served as important sources of therapeutic compounds and continue to play a vital role in healthcare systems, particularly in developing countries. The World Health Organization estimates that a large proportion of the global population relies on traditional medicine derived from medicinal plants for primary healthcare needs (WHO, 2014). Numerous plant-derived compounds have been successfully incorporated into modern medicine, and approximately one-quarter of currently available pharmaceutical products are directly or indirectly derived from plant sources (Chin et al., 2006). Therefore, medicinal plants remain valuable reservoirs of bioactive molecules with significant pharmaceutical potential.

Among medicinal plants, *Psidium guajava* L. (guava) has attracted considerable attention because of its broad range of pharmacological properties. Belonging to the family Myrtaceae, guava is widely distributed in tropical and subtropical regions and has been traditionally used for the treatment of diarrhoea, wounds, ulcers, gastrointestinal disorders, and infectious diseases (Begum et al., 2002; Dakappa et al., 2013). The leaves

are particularly rich in flavonoids, tannins, polyphenols, and quercetin derivatives, compounds known for their antimicrobial, antioxidant, anti-inflammatory, antiviral, and antitumor activities (Daswani et al., 2017). Guava extracts have demonstrated inhibitory activity against several pathogenic bacteria and fungi, supporting their traditional medicinal use (Kubmarawa et al., 2007; Kumar et al., 2021).

Similarly, *Mangifera indica* L. (mango), a member of the family Anacardiaceae, is recognized not only as an economically important fruit crop but also as a valuable medicinal plant. Various parts of the mango tree, including leaves, bark, seeds, and kernels, contain bioactive compounds such as mangiferin, flavonoids, tannins, phenolic acids, and terpenoids (Barreto et al., 2008; Batool et al., 2018). These compounds exhibit diverse biological activities, including antimicrobial, antioxidant, anti-inflammatory, antidiabetic, and anticancer properties (Salem et al., 2013; Mirza et al., 2021). Mangiferin, the principal bioactive constituent of mango leaves, has been reported to possess strong antimicrobial activity against several bacterial pathogens (Ouf et al., 2021).

Plant-derived phytochemicals have emerged as promising alternatives to conventional antibiotics because they can inhibit microbial growth through multiple mechanisms, thereby reducing the likelihood of resistance development (Dzotam and Kuete, 2017; Khameneh et al., 2021). Several studies have demonstrated the effectiveness of guava and mango leaf extracts against clinically important pathogens, including *Escherichia coli, Staphylococcus aureus*, *Salmonella typhi*, *Pseudomonas aeruginosa*, *Candida albicans, and Mycobacterium tuberculosis* (Dakappa et al., 2013; Bharti, 2013; Ouf et al., 2021). Furthermore, Manzur et al. (2020) reported significant antimicrobial and antibiofilm activity of mango leaf extracts against *Staphylococcus aureus* isolated from bovine mastitis cases.

The increasing prevalence of MDR bacterial strains and the declining effectiveness of conventional antibiotics highlight the necessity of exploring novel antimicrobial agents from natural sources. Medicinal plants such as *Psidium guajava* and *Mangifera indica* provide an attractive avenue for developing alternative antimicrobial therapies because of their rich phytochemical composition, broad-spectrum biological activities, and long history of traditional use. Therefore, the present study was undertaken to evaluate the antimicrobial activity of *Mangifera indica* and *Psidium guajava* leaf extracts against selected antibiotic-resistant bacterial strains and to assess their potential as natural alternatives for combating antimicrobial resistance.

MATERIALS AND METHODS

Collection and Identification of Plant Materials

Fresh leaves of *Mangifera indica* L. and *Psidium guajava* L. were collected from the vicinity of the Department of Microbiology, University of Ibadan, Ibadan, Nigeria. The collected plant materials were authenticated at the Herbarium Unit, Department of Botany, University of Ibadan, where voucher specimens were deposited for future reference.

Preparation of Plant Extracts

The collected leaves were thoroughly washed with distilled water to remove dust and other contaminants, air-dried at room temperature, and ground into a fine powder using an electric laboratory blender. One hundred grams of the powdered material was separately macerated in 1 L of methanol, ethanol, and distilled water in sterile conical flasks for 72 h at ambient temperature with intermittent shaking. Extraction was carried out following the method of Akerele et al. (2008) with slight modifications. The mixtures were filtered through Whatman No. 1 filter paper, and the filtrates were concentrated under reduced pressure at 40°C using a rotary evaporator (Heidolph, Germany). The concentrated extracts were transferred into sterile airtight containers and stored at 4°C until further analysis. Extraction yield was calculated using

Extraction yield (%)= (Extract weight/ dry plant weight) x 100

Collection and Identification of Test Organisms

Clinical isolates of *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Klebsiella pneumoniae* were obtained from the Department of Microbiology, University of Ibadan. Pure cultures were obtained by sub-culturing on nutrient agar and incubating for 24 h. The bacterial isolates were identified using standard morphological and biochemical characterization procedures as described by Cheesbrough (2018) and confirmed as multidrug-resistant (MDR) by Kirby–Bauer disc diffusion on Mueller–Hinton agar (MHA) according to CLSI guidelines.

Antimicrobial Susceptibility Assay

Antibacterial activity of the plant extracts was evaluated using the agar well diffusion method. Fresh bacterial suspensions were prepared in sterile normal saline and adjusted to a 0.5 McFarland turbidity standard (approximately 1.5×10^8 CFU mL^{-1}). Sterile cotton swabs were used to uniformly inoculate Mueller–Hinton Agar (MHA) plates. Wells measuring 6 mm in diameter and 5 mm in depth were aseptically bored into the agar using a sterile cork borer. Three concentrations (100 g/mL, 50 g/mL and 25 g/mL) of each extract were prepared, and 0.5 mL of each concentration was dispensed into the respective wells. Gentamicin served as the positive control, while dimethyl sulfoxide (DMSO) was used as the negative control. Plates were incubated at 37°C for 18–24 h, after which zones of inhibition were measured in millimetres (mm).

Determination of Minimum Inhibitory Concentration (MIC)

The MIC of each extract was determined using the broth dilution method described by Ali et al. (2017). Stock solutions of the extracts (100 g mL^{-1}) were subjected to two-fold serial dilution in nutrient broth to obtain

concentrations of 100, 50, 25, 12.5, 6.25, and 3.125 mg mL^{-1}. Each tube was inoculated with 50 µL of standardized bacterial suspension equivalent to a 0.5 McFarland standard and incubated at 37°C for 24 h. Control tubes containing extract and broth without bacterial inoculum were included. The MIC was recorded as the lowest extract concentration that showed no visible bacterial growth.

Determination of Minimum Bactericidal Concentration (MBC)

Aliquots (0.1 mL) from MIC tubes showing no visible growth were streaked onto sterile Mueller–Hinton Agar plates and incubated at 37°C for 48 h. The MBC was defined as the lowest concentration of extract that completely inhibited bacterial growth on the agar plates.

Qualitative Phytochemical Screening

Qualitative phytochemical screening of the extracts was conducted using standard procedures described by Harborne (1998) and Sofowora (2008). The presence or absence of major phytochemical constituents, including alkaloids, flavonoids, tannins, saponins, phenolics, and terpenoids, was determined.

Quantitative Phytochemical Analysis

Quantitative determination of total alkaloids, flavonoids, tannins, saponins, phenolics, and terpenoids was carried out according to the methods described by AOAC (2019). Results were expressed as percentage (%) composition of dry extract weight.

Statistical Analysis

All experiments were conducted in duplicate, and data were expressed as mean ± standard deviation (SD). Statistical analyses were performed using SPSS version 25.0 (IBM Corp., Armonk, NY, USA). Differences among treatment means were evaluated using one-way analysis of variance (ANOVA), and significant differences were separated using Duncan's Multiple Range Test (DMRT) at a 5% probability level ($p \leq 0.05$).

RESULTS AND DISCUSSION

Extraction Yield and Phytochemical Screening

Methanol extracts yielded the highest crude material from both species: *M. indica* (12.3 ± 0.4%) and *P. guajava* (14.8 ± 0.6%) w/w, followed by ethanol (8.7 ± 0.3%; 9.5 ± 0.4%) and aqueous extracts (6.2 ± 0.5%; 5.9 ± 0.3%), with significant differences across solvents ($p < 0.05$). Methanol's intermediate polarity enables co-extraction of both polar (glycosides, alkaloids) and moderately non-polar (terpenoids, aglycone flavonoids) metabolites simultaneously, explaining the superior yield and the more comprehensive qualitative profiles observed in Table 1.

The results in Table 1 indicate a relatively high abundance of saponins, tannins, flavonoids, and cardiac glycosides across all solvent types. Terpenoids were moderately present in aqueous and ethanol extracts, but were abundantly present in the methanolic extracts. Steroids were absent in the aqueous extracts, and are abundant in both ethanol and methanol. Anthraquinones were detected only in the methanol extracts, moderately present in aqueous extracts, and absent in ethanol. Tables 2 and 3 present the quantitative analysis of phytochemicals in methanol, ethanol, and aqueous extracts of *Mangifera indica* and *Psidium guajava* leaves. *M. indica* showed the highest alkaloid content in the methanol extract (11.40% w/w), followed by aqueous (9.45%) and ethanol (8.65%), while *P. guajava* exhibited even higher alkaloid levels, with methanol extract containing 17.20%, ethanol 10.40%, and aqueous 10.00%. Similarly, the highest value of flavonoids were recorded in methanol extracts of *M. indica* and *P. guajava* had the highest contents (0.59% and 5.12%, respectively). *M. indica* had little yield of saponin in methanol (2.00%), while *P. guajava* had the highest in ethanol (9.40%). Tannin content was relatively stable across solvents for both species. Terpenoids were highest in methanol for *M. indica* (1.35%) and in ethanol for *P. guajava* (1.70%). Phenol content showed variation across extracts; however, aqueous extracts of *M. indica* had 0.82%, while 0.98% were recorded in *P. guajava* methanol extract.

Antimicrobial Susceptibility Testing

The antimicrobial activity of *Mangifera indica* and *Psidium guajava* leaf extracts was assessed using aqueous, ethanol, and methanol solvents at concentrations of 100, 50, and 25 g/mL against five bacterial isolates: *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, and *Staphylococcus epidermidis* (Tables 4 and 5). In both species, methanol extracts demonstrated the strongest and most consistent antibacterial activity, particularly at 100 g/mL, where inhibition zones exceeded 23 mm against *S. aureus* and *E. coli*., as shown in Fig. 1a and 1b. Ethanol extracts also showed moderate efficacy, especially against *S. aureus* and *P. aeruginosa*, while aqueous extracts were generally less active but still produced notable inhibition, particularly against *S. aureus* (up to 26 mm in *M. indica*) as shown in Fig. 1a and 1c. Across all extracts and concentrations, *S. aureus* was the most susceptible organism, followed by *P. aeruginosa* and *S. epidermidis*. Also, *K. pneumoniae* exhibited the least susceptibility, with inhibition zones consistently below 15 mm as shown in Fig. 1d. A general decline in antibacterial activity was observed with decreasing extract concentration.

Minimum Inhibitory Concentration (MIC)

The MIC results for *Mangifera indica* and *Psidium guajava* extracts against five bacterial isolates are summarised in Tables 6 and 8. Across both species, methanol extracts consistently demonstrated the lowest MIC values, indicating the strongest antimicrobial activity, followed by ethanol and aqueous extracts. For *E. coli* and *S. aureus*, methanol extracts of both plants were most effective, with MIC values of 12.5 g/mL in *M. indica* and inhibition observed down to 12.5% in *P. guajava*. *P. aeruginosa* was highly susceptible to ethanol and methanol extracts from *P. guajava*, showing inhibition at concentrations as low as 6.25%, while *M. indica* extracts were effective at 25 g/mL across all

solvents. *S. epidermidis* showed the greatest susceptibility to ethanol (*M. indica*, 12.5 g/mL) and methanol (*P. guajava*, 6.25%) extracts. Also, *K. pneumoniae* exhibited higher MIC value, with most extracts requiring 25 g/mL or higher to inhibit growth.

Minimum Bactericidal Concentration (MBC)

Tables 7 and 9 shows the MBC values of aqueous, ethanol, and methanol extracts of *Mangifera indica* and *Psidium guajava* against five bacterial isolates. Methanol and ethanol extracts of both plants generally showed stronger bactericidal activity than aqueous extracts, with *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Staphylococcus epidermidis* being the most susceptible. In *M. indica*, methanol extract exhibited the lowest MBC (12.5 g/mL) against *S. aureus* and *S. epidermidis*, while aqueous and ethanol extracts showed moderate activity (25–50 g/mL). For *E. coli* and *K. pneumoniae*, MBCs ranged from 25–50 g/mL across all extracts, indicating lower susceptibility. *P. aeruginosa* showed moderate sensitivity, with MBCs between 25–50 g/mL depending on the extract.In *P. guajava*, methanol and ethanol extracts were more bactericidal at lower concentrations. Notably, *P. aeruginosa* was completely eradicated at 3.125% for both methanol and ethanol extracts, and *S. epidermidis* at 6.25% with methanol. *S. aureus* responded to ethanol at 6.25% and methanol at 12.5%, while aqueous extracts were only effective at higher concentrations (25–50%). *K. pneumoniae* was least responsive, with ethanol failing to show bactericidal activity even at 50%.

Table 1. Qualitative phytochemical screening of *Mangifera indica* and *Psidium guajava* leaf extracts

	Mangifera indica leaf extracts			*Psidium guajava* leaf extracts		
Compound	Aqueous	Ethanol	Methanol	Aqueous	Ethanol	Methanol
Saponins	++	++	++	++	++	++
Tannins	++	++	++	++	++	++
Flavonoids	++	++	++	++	++	++
Cardiacglycosides	++	++	++	++	+	++
Terpenoids	+	+	++	+	+	++
Steroids	-	++	++	-	++	++
Anthraquinones	+	-	++	+	++	+
Phenol	+	++	++	+	++	++
Alkaloids	++	++	++	++	++	++

++ = Strongly positive, + = Positive, - = Negative

Table 2. Quantitative phytochemical screening of *Mangifera indica* leaf extracts

Sample	Alkaloids (%w/w)	Flavonoids (%w/w)	Saponins (%w/w)	Tannin (%w/w)	Terpenoids (%w/w)	Phenol (%w/w)
Methanol extract	11.40±0.004	0.59±0.063	2.00±0.003	1.29±0.010	1.35±0.004	0.78±0.006
Ethanol extract	8.65±0.004	0.37±0.180	0.70±0.003	1.14±0.005	1.00±0.003	0.78±0.004
Aqueous extract	9.45±0.004	0.20±0.149	1.50±0.003	1.25±0.003	0.60±0.001	0.82±0.005

Table 3: Quantitative phytochemical screening of *Psidium guajava* leaf extracts

Sample	Alkaloids (%w/w)	Flavonoids (%w/w)	Saponins (%w/w)	Tannin (%w/w)	Terpenoids (%w/w)	Phenol (%w/w)
Methanol extract	17.20±0.003	5.12±0.109	7.50±0.003	1.48±0.149	1.50±0.004	0.98±0.004
Ethanol extract	10.40±0.003	3.22±0.109	9.40±0.003	1.46±0.005	1.70±0.003	0.79±0.003
Aqueous extract	10.00±0.001	0.94±0.123	0.70±0.003	1.39±0.009	0.80±0.001	0.72±0.007

Key: Mean ± Standard deviation of duplicate measurements of the zone of inhibition

Table 4. Zones of inhibition of Aqueous, Ethanolic and Methanolic leaf extracts of *Psidium guajava*

	Extracts								
	Aqueous extract (g/mL)			**Ethanol extract (g/mL)**			**Methanol extract (g/mL)**		
Microorganism	**100**	**50**	**25**	**100**	**50**	**25**	**100**	**50**	**25**
Pseudomonas aeruginosa	16.5 ±0.98	15.5 ± 2.94	12 ±0.00	20 ± 0.00	13 ±0.00	11.5 ±0.98	22.5 ±0.98	15 ±0.00	12.5 ±0.98
Escherichia coli	14.5 ±0.98	11.5 ± 0.98	11 ±0.00	15.5 ±0.98	11 ±0.00	10.5 ±0.98	25 ±1.96	13 ±0.00	11 ±1.96
Staphylococcus aureus	22.5 ±0.98	23 ± 1.96	20 ±0.00	22.5 ±0.98	19 ±0.00	16 ±0.00	24.5 ±0.98	21. ±0.98	20.25 ±0.49
Klebsiella pneumoniae	15 ±0.00	13 ± 3.92	10 ±0.00	12.5 ±0.98	13 ±0.00	11 ±0.00	14.5 ±0.98	14.5±0.98	10.5 ±0.98
Staphylococcus epidermidis	12 ±0.00	12 ± 1.96	13.5 ±2.94	20 ±0.00	20 ±1.96	16.5 ±0.98	21 ±1.96	15 ±3.92	12.5 ±0.98

Key: Mean ± standard deviation of duplicate measurements of the zone of inhibition

Table 5. Zones of inhibition of Aqueous, Ethanolic and Methanolic leaf extracts of *Mangifera indica*

	Extracts								
	Aqueous (g/mL)			**Ethanol (g/mL)**			**Methanol (g/mL)**		
Isolates	**100**	**50**	**25**	**100**	**50**	**25**	**100**	**50**	**25**
Pseudomonas aeruginosa	17 ±0.00	14.5±0.98	12±0.00	14.5±0.98	13.5±2.94	12.5±0.98	23.5±0.98	15 ±0.00	13±1.96
Staphylococcus aureus	26 ±0.00	24.5±0.98	20 ±0.00	16.5±4.9	15 ±1.96	14 ±3.92	25.5±0.98	20 ±0.00	19±0.00
Escherichia coli	12.5 ±0.98	12.5±0.00	12 ±0.00	16 ±1.96	13 ±1.96	11 ±1.96	18.5±2.94	12±0.00	11±1.96
Klebsiella pneumonia	17.5±0.98	16.5±0.98	12 ±0.00	14 ±1.96	10.5±0.98	10 ±0.00	13.5±2.94	13.5±0.98	10.5±0.98
Staphylococcus epidermidis	17.5±0.98	14 ±0.00	13±1.96	21 ±0.00	15.5 ±0.98	15±0.00	23 ±0.00	18.5±0.98	15 ±0.00

Key: Mean ± Standard deviation of duplicate measurements of the zone of inhibition

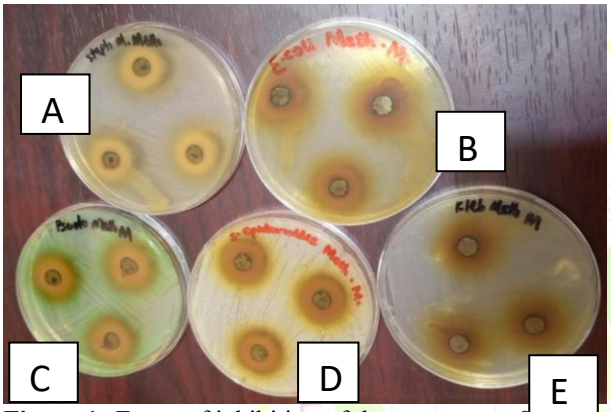

Figure 1: Zones of inhibition of the extracts on *S. aureus* (A), *E. coli* (B), *Pseudomonas aeruginosa* (C), *K. pneumonia*(E), *S. epidermidis*(D)

Table 6. Minimum Inhibitory Concentration of the *Mangifera indica* leaf extracts against bacterial isolates

Concentrations ((g/mL)		50	25	12.5	6.25	3.125	MIC
Organism	Extract						
Escherichia coli	Aqueous	-	-	+	+	+	25
	Ethanol	-	+	+	+	+	50
	Methanol	-	-	-	+	+	12.5
Klebsiella pneumoniae	Aqueous	-	-	+	+	+	25
	Ethanol	-	+	+	+	+	50
	Methanol	-	+	+	+	+	50
Staphylococcus aureus	Aqueous	-	+	+	+	+	50
	Ethanol	-	-	+	+	+	25
	Methanol	-	-	-	+	+	12.5
Pseudomonas aeruginosa	Aqueous	-	-	+	+	+	25
	Ethanol	-	-	+	+	+	25
	Methanol	-	-	+	+	+	25
Staphylococcus epidermidis	Aqueous	-	-	+	+	+	25
	Ethanol	-	-	-	+	+	12.5
	Methanol	-	-	+	+	+	25

+ = Growth - = No growth

Table 7: Minimum Bactericidal Concentration (MBC) results of leaf extracts of *Mangifera indica* against bacterial isolates

Concentrations	(g/mL)	50	25	12.5	6.25	3.125	MBC
Organism	Extract						
Escherichia Coli	Aqueous	-	-	+	+	+	25
	Ethanol	-	+	+	+	+	50
	Methanol	-	+	+	+	+	50
Klebsiella pneumoniae	Aqueous	-	+	+	+	+	50
	Ethanol	-	+	+	+	+	50
	Methanol	-	+	+	+	+	50
Staphylococcus aureus	Aqueous	-	-	+	+	+	25
	Ethanol	-	-	+	+	+	25
	Methanol	-	-	-	+	+	12.5
Pseudomonas aeruginosa	Aqueous	-	-	+	+	+	25
	Ethanol	-	+	+	+	+	50
	Methanol	-	-	+	+	+	25
Staphylococcus epidermidis	Aqueous	-	-	-	+	+	12.5
	Ethanol	-	+	+	+	+	50
	Methanol	-	-	-	+	+	12.5

+ = Growth - = No growth

Table 8. Minimum Inhibitory Concentration of the three Extracts of *Psidium guajava* against Bacterial Isolates

		Concentrations (g/mL)				
Microorganism	Extract	50	25	12.5	6.25	3.125
Escherichia coli	Aqueous	-	-	+	+	+
	Ethanol	-	-	+	+	+
	Methanol	-	-	+	+	+
Pseudomonas aeruginosa	Aqueous	-	-	+	+	+
	Ethanol	-	-	-	-	+
	Methanol	-	-	-	-	+
Klebsiella pneumoniae	Aqueous	-	-	+	+	+
	Ethanol	-	-	+	+	+
	Methanol	-	-	+	+	+
Staphylococcus aureus	Aqueous	-	+	+	+	+
	Ethanol	-	-	-	+	+
	Methanol	-	-	+	+	+
Staphylococcus epidermidis	Aqueous	-	-	+	+	+
	Ethanol	-	-	-	+	+
	Methanol	-	-	-	-	+

Key: +: indicates growth; -: indicates no growth

Table 9. Minimum Bactericidal Concentration (MBC) results of different extracts of *Psidium guajava* against bacterial isolate

		Concentrations (g/mL)				
Microorganism	Extract	50	25	12.5	6.25	3.125
Escherichia coli	Aqueous	-	+	+	+	+
	Ethanol	-	+	+	+	+
	Methanol	-	+	+	+	+
Pseudomonas aeruginosa	Aqueous	-	-	-	+	+
	Ethanol	-	-	-	-	+
	Methanol	-	-	-	+	+
Klebsiella pneumoniae	Aqueous	-	+	+	+	+
	Ethanol	+	+	+	+	+
	Methanol	-	+	+	+	+
Staphylococcus aureus	Aqueous	-	-	-	+	+
	Ethanol	-	-	-	+	+
	Methanol	-	-	+	+	+
Staphylococcus epidermidis	Aqueous	-	-	-	+	+
	Ethanol	-	-	+	-	+
	Methanol	-	-	-	-	+

+: indicates growth ;- : indicates no growth

The global rise of multidrug-resistant (MDR) bacteria has renewed interest in plant-derived antimicrobials as potential alternatives or complements to conventional antibiotics. In the present study, leaf extracts of *Mangifera indica* and *Psidium guajava* exhibited notable antibacterial activities against clinically important bacterial pathogens, thereby validating their traditional medicinal applications and corroborating previous reports on their antimicrobial efficacy (Bharti, 2013; Manzur et al., 2020). Phytochemical screening revealed the presence of several bioactive constituents, including alkaloids, flavonoids, tannins, saponins, terpenoids, steroids, phenols, and anthraquinones. These phytochemicals are known to possess antimicrobial properties through diverse mechanisms such as disruption of cell membrane integrity, inhibition of cell wall synthesis, interference with nucleic acid replication, and suppression of protein biosynthesis (Cowan, 1999; Nijveldt et al., 2001; Khameneh et al., 2021). Among the extraction solvents evaluated, methanol extracts contained the highest diversity and concentration of phytochemicals, which was reflected in their superior antibacterial activity. This finding is consistent with earlier studies reporting that organic solvents are generally more efficient than water in extracting secondary metabolites from medicinal plants (Dakappa et al., 2013; Ouf et al., 2021).

The antimicrobial susceptibility assay demonstrated that both plant species were more effective against Gram-positive bacteria, particularly *Staphylococcus aureus* and *Staphylococcus epidermidis*, than against Gram-negative pathogens such as *Escherichia coli*, *Pseudomonas aeruginosa*, and *Klebsiella pneumoniae*. This differential susceptibility may be attributed to structural differences in bacterial cell walls. The outer lipopolysaccharide membrane of Gram-negative bacteria acts as a permeability barrier that limits the penetration of many antimicrobial compounds (Delcour, 2009; Khameneh et al., 2021). Among the tested organisms, *S. aureus* consistently exhibited the largest inhibition zones, suggesting high sensitivity to the plant extracts. This observation is particularly important given the increasing prevalence of antibiotic-resistant Staphylococcus strains in clinical settings (Tong et al., 2015).

The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) results further supported the agar diffusion assay findings. Methanolic extracts showed the lowest inhibitory and bactericidal concentrations, especially against *S. aureus*, *S. epidermidis*, and *P. aeruginosa*. Similar findings have been reported by Bharti (2013), Dakappa et al. (2013), and Ouf et al. (2021), who observed strong antibacterial activity of methanol and ethanol extracts of *M. indica* and *P. guajava* against MDR pathogens. The comparatively lower efficacy of aqueous extracts highlights the importance of solvent selection for maximizing the extraction of bioactive compounds.

Particularly noteworthy was the strong bactericidal activity of *P. guajava* methanol and ethanol extracts against *P. aeruginosa* and *S. epidermidis* at relatively low concentrations. Since *P. aeruginosa* possesses multiple intrinsic and acquired resistance mechanisms, including efflux pumps and biofilm formation, effective inhibition of this pathogen by guava extracts suggests significant therapeutic potential (Jairath et al., 2021). Consequently, *P. guajava* may represent a promising source of natural antimicrobial compounds for the development of adjunct therapies against difficult-to-treat bacterial infections.

Overall, the findings indicate that *M. indica* and *P.*

guajava leaves contain diverse phytochemicals that exert both bacteriostatic and bactericidal effects. The presence of multiple active compounds may reduce the likelihood of resistance development compared with conventional single-target antibiotics (Wagner and Ulrich-Merzenich, 2009; Ruddaraju et al., 2020). Nevertheless, the present study was limited to in vitro evaluation. Further research involving bioactive compound isolation, toxicity assessment, pharmacological characterization, in vivo validation, and synergistic studies with conventional antibiotics is necessary before clinical application can be recommended.

CONCLUSION

This study demonstrated that *extracts of Mangifera indica and Psidium guajava*, particularly methanol extracts, exhibit higher antimicrobial activity against antibiotic-resistant bacterial strains. The phytochemical analysis confirmed the presence of bioactive compounds responsible for these effects, emphasizing the potential of the extracts as an alternative treatment option for bacterial infections. The effectiveness of these extracts against Gram-positive and some Gram-negative bacteria used in this study suggests their potential use in managing antibiotic-resistant infections.

Funding- No grant was received for this research.
Informed Consent statement- Not applicable
Data Availability Statement- Not applicable
Acknowledgement- The authors appreciate to Mr. Fadiora, Chief technologist and other laboratory technicians of Department of Microbiology, University of Ibadan, Ibadan, Oyo State for their constructive contributions.
Conflicts of Interest- No conflict of interest was declared.
Author Contributions- J.O.A., O.J.J. and A.A.A.: Conceptualization; J.O.A., O.M.A., and O.J.J.; Methodology, data curation; O.J.J.: Software; J.O.A. and A.A.A.: validation; J.O.A: formal analysis; H.E.A. and O.M.A.: investigation, resources; H.E.A. and O.M.A. and A.A.A.: Writing- original draft preparation; J.O.A., O.J.J. and A.A.A.: Writing- review and editing; J.O.A., and O.J.J.: visualization; J.O.A.: supervision, project administration. All authors have read and agreed to the published version of the manuscript.

REFERENCES

Akerele, J.O., Obasuyi, O. and Ewhre, L.O., 2008. Antimicrobial activity of the extracts of Dialium guineense (Willd) stem bark. *Journal of Applied Sciences*, **11**(16): 2923–2928.

Ali, S.M., Khan, N.A. and Sagathevan, K., 2017. In vitro antifungal activity of bioactive compounds from Allium sativum and Piper betle against azole-resistant Candida albicans. *Journal of Deployed Sciences*, 1(1), 1-6.

AOAC International, 2019. Official Methods of Analysis of AOAC International, 21st ed. Gaithersburg, MD: AOAC International.

Barreto, J.C., Trevisan, M.T.S., Hull, W.E., Erben, G., de Brito, E.S., Pfundstein, B., Würtele, G., Spiegelhalder, B. and Owen, R.W., 2008. Characterisation and quantitation of polyphenolic compounds in bark, kernel, leaves, and peel of mango (*Mangifera indica* L.). *Journal of Agricultural and Food Chemistry*, **56**(14): 5599–5610. https://doi.org/10.1021/jf800738r

Batool, N., Ilyas, N., Shabir, S., Saeed, M. and Mazhar, R., 2018. A mini-review of the therapeutic potential of *Mangifera indica* L. *Pakistan Journal of Pharmaceutical Sciences*, **31**(4): 1441–1448.

Bharti, S., 2013. Mangifera indica (Mango): A promising medicinal plant for breast cancer therapy and prevention of carcinogenesis. *Journal of Cancer Therapy,* **4**(4): 872–878.

Begum, S., Hassan, S.I. and Siddiqui, B.S., 2002. Two new triterpenoids from the fresh leaves of *Psidium guajava*. *Planta Medica*, **68**: 1149–1152. https://doi.org/10.1055/s-2002-36363

Cheesbrough, M., 2006 (reprinted 2018). District Laboratory Practice in Tropical Countries, Part 2, 2nd ed. Cambridge: Cambridge University Press.

Cowan, M.M., 1999. Plant Products as Antimicrobial Agents. *Clinical Microbiology Reviews*, **12**(4): 564–582. https://doi.org/10.1128/cmr.12.4.564

Chin, Y.W., Balunas, M.J., Chai, H.B. and Kinghorn, A.D., 2006. Drug discovery from natural sources. *The AAPS Journal*, **8**(2): E239–E253. https://doi.org/10.1208/aapsj080229

Dakappa, S.S., Adhikari, R., Timilsina, S.S. and Sajjekhan, S., 2013. A review on the medicinal plant *Psidium guajava* Linn. (Myrtaceae). *Journal of Drug Delivery and Therapeutics*, **3**(2): 162–168.

Daswani, P.G., Gholkar, M.S. and Birdi, T.J., 2017. *Psidium guajava*: A single plant for multiple health problems of the rural Indian population. *Pharmacognosy Reviews*, **11**(22): 167–174. https://doi.org/10.4103/phrev.phrev_45_16

Delcour, A.H., 2009. Outer membrane permeability and antibiotic resistance. *Biochimica et Biophysica Acta (BBA) - Proteins and Proteomics*, **1794**(5): 808–816. https://doi.org/10.1016/j.bbapap.2008.11.005

Dzotam, J.K. and Kuete, V., 2017. Antibacterial and antibiotic-modifying activity of methanol extracts from six Cameroonian food plants against multidrug-resistant enteric bacteria. *BioMed Research International*, **2017**: 1583510. https://doi.org/10.1155/2017/1583510

Ficker, C.E., Arnason, J.T., Vindas, P.S., Alvarez, L.P., Akpagana, K., Gbeassor, M., De Souza, C. and Smith, M.L., 2003. Inhibition of human pathogenic fungi by ethnobotanically selected plant extracts. *Mycoses*, **46**(1–2): 29–37.

Harborne, J.B., 1998. Phytochemical Methods: A Guide to Modern Techniques of Plant Analysis, 3rd ed. London: Chapman and Hall.

Hughes, D. and Andersson, D.I., 2017. Evolutionary trajectories to antibiotic resistance. *Annual Review of Microbiology*, **71**: 579–596. https://doi.org/10.1146/annurev-micro-090816-093813

Jairath, G., Singh, P.K. and Shukla, S., 2021. Biofilm formation and antimicrobial resistance in *Pseudomonas aeruginosa*: A review. *Journal of Global Antimicrobial Resistance*, **26**: 225–230.

Kubmarawa, D., Ajoku, G.A., Enwerem, N.M. and Okorie, D.A., 2007. Preliminary phytochemical and antimicrobial screening of 50 medicinal plants from Nigeria. African Journal of Biotechnology, 6(14): 1690–1696.

Khameneh, B., Iranshahy, M., Soheili, V. and Bazzaz, B.S.F., 2021. Review on plant antimicrobials: A mechanistic viewpoint. *Antimicrobial Resistance and Infection Control*, **10**: 118. https://doi.org/10.1186/s13756-021-00979-8

Kumar, M., Prakash, S., Kumari, N., Pundir, A. and Punia, S., 2021. Phytochemicals as antimicrobial agents: A review. *Current Research in Food Science*, **4**: 672–679. https://doi.org/10.1016/j.crfs.2021.09.002

Lewis, K. and Ausubel, F.M., 2006. Prospects for plant-derived antibacterials. *Nature Biotechnology*, **24**(12): 1504–1507. https://doi.org/10.1038/nbt1206-1504

Manzur, A.G.B., Junior, V.S.M., Morais-Costa, F., Mariano, E.G.A., Careli, R.T., da Silva, L.M.V., Coelho, S.G., de Almeida, A.C. and Duarte, E.R., 2020. Extract of Mangifera indica L. leaves may reduce biofilms of Staphylococcus spp. in stainless steel and teatcup rubbers. Food Science and Technology International, 26(1): 3–12. https://doi.org/10.1177/1082013219858529

Mirza, M.B., Sastry, V.G. and Khan, M.A., 2021. Medicinal and therapeutic potential of *Mangifera indica*: A review. *Journal of Pharmacognosy and Phytochemistry*, **10**(1): 1223–1229.

Morar, M. and Wright, G.D., 2010. The genomic enzymology of antibiotic resistance. *Annual Review of Genetics*, **44**: 25–51. https://doi.org/10.1146/annurev-genet-102209-163517

Nijveldt, R.J., van Nood, E., van Hoorn, D.E.C., Boelens, P.G., van Norren, K. and van Leeuwen, P.A.M., 2001. Flavonoids: A review of probable mechanisms of action and potential applications. *The American Journal of Clinical Nutrition*, **74**(4): 418–425. https://doi.org/10.1093/ajcn/74.4.418

Ouf, S.A., Moussa, T.A. and Abdel-Meguid, E.M., 2021. Anti-mycobacterial activity of *Mangifera indica* L. leaves against *Mycobacterium tuberculosis*. *Journal of Applied Pharmaceutical Science*, **11**(4): 108–114.

Ruddaraju, L.K., Pammi, S.V.N. and Gadi, S., 2020. A review on antibiotics and plant-based antimicrobials to combat antibiotic resistance. *International Journal of Pharmaceutical Sciences and Research*, **11**(3): 1092–1105.

Sofowora, A., 2008. Medicinal Plants and Traditional Medicine in Africa, 3rd ed. Ibadan: Spectrum Books Limited.

Tong, S.Y.C., Davis, J.S., Eichenberger, E., Holland, T.L. and Fowler, V.G., 2015. Staphylococcus aureus infections: epidemiology, pathophysiology, clinical manifestations, and management. Clinical Microbiology Reviews, 28(3): 603–661. https://doi.org/10.1128/CMR.00134-14

Salem, M.Z.M., Ali, H.M. and El-Shanhorey, N.A., 2013. Evaluation of the antibacterial activity of *Mangifera indica* L. leaves and stem extracts against some pathogenic bacteria. *Journal of Medicinal Plants Research*, **7**(12): 746–752.

World Health Organization, 2014. Antimicrobial Resistance: Global Report on Surveillance. Geneva: WHO Press.

Wagner, H. and Ulrich-Merzenich, G., 2009. Synergy research: Approaching a new generation of phytopharmaceuticals. *Phytomedicine*, **16**(2–3): 97–110. https://doi.org/10.1016/j.phymed.2008.12.018

Zilberberg, M.D., Nathanson, B.H., Sulham, K., Fan, W. and Shorr, A.F., 2016. Multidrug resistance, inappropriate empiric therapy, and hospital mortality in Acinetobacter baumannii pneumonia and sepsis. Critical Care, 20: 221. https://doi.org/10.1186/s13054-016-1392-4

Citation: Ayoade, J.O., Joseph, O.J., Aroyehun, H.E., Adekunle, O.M. and Adegoke, A.A., 2026. In vitro antimicrobial activities of leaf extracts of *Mangifera indica* Dehnh. and *Psidium guajava* F. Muell. against selected antibiotic-resistant bacteria strains. *International Journal of Agricultural and Applied Sciences*, 7(1): 55-62**.** https://doi.org/10.52804/ijaas2026.716