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## Research Article

# Antibacterial Activity of *Curcuma mangga* Extracts Against Antibiotic-Resistant Bacteria: Natural Alternative for Combating Multidrug Resistance

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## Abstract

**Background and Objective:** *Curcuma mangga*, commonly known as mango ginger, is a medicinal plant renowned for its anti-inflammatory, antioxidant and digestive health benefits, attributed to its high content of curcuminoids and essential oils. This study aims to assess the antibacterial potential of *C. mangga* extracts against five antibiotic-resistant bacterial strains and five standard pathogenic reference strains. **Materials and Methods:** *C. mangga* rhizomes were dried, ground into powder and individually extracted using ethanol, dichloromethane and hexane. The antibacterial activity was assessed using the microbroth dilution method to find the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC). Statistical analysis was performed using Duncan's Multiple Range Test (DMRT) to show significant differences among mean values at a 95% confidence level. **Results:** The dichloromethane and hexane extracts exhibited the strongest inhibitory effect, showing the lowest MIC value of 0.049 mg/mL against multidrug-resistant *Klebsiella pneumoniae* and *Stenotrophomonas maltophilia*. The dichloromethane extract showed the strongest bactericidal effect against *S. maltophilia*, with the lowest MBC value of 1.56 mg/mL. **Conclusion:** This study is the first to prove that *C. mangga* extracts exhibit significant antibacterial and bactericidal activity against several antibiotic-resistant pathogens, including multidrug-resistant *K. pneumoniae* and *S. maltophilia*. These findings highlight the potential of *C. mangga* as a natural antimicrobial agent and lay the groundwork for the development of plant-based alternatives to address the growing threat of antibiotic resistance.

**Key words:** *Curcuma mangga* extracts, antibacterial activity, multidrug-resistant *Klebsiella pneumoniae*, *Stenotrophomonas maltophilia*, curcuminoids

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**Competing Interest:** The authors have declared that no competing interest exists.

**Data Availability:** All relevant data are within the paper and its supporting information files.

## INTRODUCTION

Bacteria represent the most abundant and diverse group of microorganisms and can be broadly categorized based on their ecological niche as either intracellular or extracellular organisms<sup>1</sup>. While a significant proportion of bacterial species contribute beneficially to human health, particularly through their roles in the gut microbiota, others are pathogenic and capable of causing infections that range from mild to life-threatening<sup>2</sup>. Pathogenic bacteria are unicellular prokaryotic organisms, typically measuring 0.5-2.0  $\mu\text{m}$  in diameter and 1-10  $\mu\text{m}$  in length and exhibiting various morphological forms such as bacilli, cocci and spirilla. These organisms are remarkably adaptable and can thrive across a wide range of environmental conditions. Due to their ability to cause disease, pathogenic bacteria have historically been managed through the use of antibiotics, however, the widespread and often inappropriate use of these drugs has led to the emergence of antibiotic-resistant strains, posing a significant global threat to public health<sup>3-5</sup>. As antibiotic resistance continues to rise at an alarming rate, the urgent need for novel and effective antimicrobial agents from alternative sources has become a global priority in infectious disease research.

The *Curcuma* genus, a member of the Zingiberaceae family, comprises numerous species known for their medicinal and culinary significance. Among them, *Curcuma mangga*, commonly known as kunyit mangga due to its mango-like aroma, is a rhizomatous herb native to Southeast Asia<sup>6</sup>. *C. mangga* is widely used in traditional medicine, dietary supplements and as a seasoning in culinary practices. Its rhizomes are traditionally employed to treat stomach disorders, fever and cancer-related conditions. Extensive pharmacological investigations of various *Curcuma* species have revealed a wide spectrum of biological activities, including anti-cancer<sup>6</sup>, antioxidant<sup>7</sup>, anti-inflammatory<sup>8</sup>, insect antifeedant, antiviral, anti-tumour<sup>9</sup>, cytotoxic, prostate disorders treatment<sup>10</sup>, antibacterial activity<sup>11</sup>, antifungal activity<sup>12</sup> and trypanocidal properties<sup>13</sup>. These therapeutic effects have been linked to their potential use in managing chronic diseases such as Alzheimer's disease and arthritis. The primary bioactive constituents responsible for these effects include curcuminoids<sup>14</sup> and a diverse array of diterpenoids<sup>15</sup>, particularly of the labdane, halimane and clerodane types.

A recent study published in 2025 reported that ethanolic extracts of *C. mangga*, at concentrations ranging from 25-200 mg/mL, showed inhibitory effects against a broad spectrum of bacterial pathogens, including *Bacillus cereus*, *Bacillus subtilis*, *Escherichia coli* and *Staphylococcus aureus*.

Remarkably, a concentration of 50 mg/mL was sufficient to exert bactericidal activity against *S. aureus*. At 100 mg/mL, the extract demonstrated bactericidal effects on both *S. aureus* and *E. coli*, highlighting its dose-dependent antimicrobial potential<sup>7</sup>. Essential oil derived from *C. mangga* exhibited the highest and broadest antimicrobial spectrum among the tested samples, effectively inhibiting the growth of all evaluated microorganisms. These included bacterial pathogens such as *B. cereus*, *S. aureus*, *E. coli* and *Pseudomonas aeruginosa*, as well as fungal species *Candida albicans* and *Cryptococcus neoformans*<sup>16</sup>. The minimum inhibitory concentration (MIC) of the ethanolic extract of *C. mangga* rhizome against *S. aureus* was determined to be 3.13%, with a minimum bactericidal concentration (MBC) of 6.25%. For *Escherichia coli*, the MIC and MBC were observed at 6.25 and 12.50%, respectively. Among the tested solvents, the ethanolic extract exhibited the strongest antibacterial activity, outperforming both the chloroform and *n*-hexane extracts<sup>17</sup>.

Given the limited data on the antibacterial efficacy of *C. mangga* rhizome extracts against antibiotic-resistant bacteria, this study aimed to evaluate their antimicrobial potential against clinically significant, drug-resistant human pathogens. The bacterial strains investigated included multidrug-resistant *Klebsiella pneumoniae*, *K. pneumoniae* strain 101, *Acinetobacter baumannii*, *Stenotrophomonas maltophilia*, *Proteus mirabilis* and colistin-resistant *Pseudomonas aeruginosa*. This research offers valuable insights into the potential of *C. mangga* rhizome extracts as a promising natural source of antibacterial agents targeting multidrug-resistant and colistin-resistant pathogens. The findings contribute to the advancement of alternative antimicrobial therapies to address the escalating global threat of antibiotic resistance.

## MATERIALS AND METHODS

**Study area:** The study was conducted between January and April, 2025 in the Microbiology Laboratory, Department of Science and Technology, Faculty of Liberal Arts and Science, Roi Et Rajabhat University, Roi Et, Thailand.

**Chemical reagent:** Ethanol, hexane and dichloromethane were purchased from Italmar (Thailand) Co., Ltd. Nutrient broth (NB) and nutrient agar (NA) were purchased from HiMedia Laboratories Pvt. Ltd. (India). Dimethyl Sulfoxide (DMSO) was procured from Sigma-Aldrich (St. Louis, Missouri, USA).



Fig.1: *Curcuma mangga* rhizomes sample

Table 1: Pathogenic bacteria

Bacterial strain	Descriptions
<i>Acinetobacter baumannii</i>	Antibiotic resistant strain, Clinical isolate
Multidrug - Resistant <i>Klebsiella pneumoniae</i>	Antibiotic resistant strain, Clinical isolate
<i>Stenotrophomonas maltophilia</i>	Antibiotic resistant strain, Clinical isolate
<i>Proteus mirabilis</i>	Antibiotic resistant strain, Clinical isolate
Colistin-resistant <i>Pseudomonas aeruginosa</i>	Antibiotic resistant strain, Clinical isolate
<i>Klebsiella pneumoniae</i> 101	Reference strain, Clinical isolate
<i>Klebsiella oxytoca</i> TISTR556	Reference strain
<i>Staphylococcus aureus</i> TISTR1466	Reference strain
<i>Staphylococcus epidermidis</i> TISTR518	Reference strain
<i>Klebsiella pneumoniae</i> TISTR1383	Reference strain

**Plant sample collection and extraction:** About 1 kg of *C. mangga* rhizomes was collected from an organic farm in Ko Kaew Subdistrict, Selaphum District, Roi Et Province, Thailand (Fig. 1). The rhizomes were thoroughly washed with tap water to remove residual soil, sliced and dried at 50°C for 48 hrs using a hot air oven (POL-EKO-APARATURA, Wodzisław Śląski, Poland). The dried rhizomes were ground into a fine powder using an herbal grinder (WF-20B THAIGRINDER, Thailand). Subsequently, 100 g of the powdered sample were separately extracted with ethanol, hexane and dichloromethane under constant agitation at 150 rpm overnight. The extracts were filtered and further dried at 50°C for 48 hrs using the hot air oven. The *C. mangga* extract was diluted in Dimethyl Sulfoxide (DMSO) to obtain a final concentration of 500 mg/mL<sup>5,18,19</sup>.

**Bacterial cultivation and preparation:** *Klebsiella oxytoca* TISTR556, *Staphylococcus aureus* TISTR1466, *Staphylococcus epidermidis* TISTR518 and *Klebsiella pneumoniae* TISTR1383

were purchased from the culture collection of the Thailand Institute of Scientific and Technological Research (TISTR), Thailand. Multidrug-resistant *Klebsiella pneumoniae*, *Klebsiella pneumoniae* 101, *Acinetobacter baumannii*, *Stenotrophomonas maltophilia*, *Proteus mirabilis* and Colistin-resistant *Pseudomonas aeruginosa* were obtained from the department of clinical microbiology, Roi Et Hospital, Roi Et, Thailand (Table 1). A single colony of each tested bacterial strain was cultured in 10 mL of nutrient broth (NB) at 37°C with continuous shaking at 150 rpm for 18 hrs. Following incubation, the bacterial cells were harvested by centrifugation and resuspended in NB to achieve a final optical density of 0.1 at 600 nm (OD) for subsequent experimental use<sup>20-22</sup>.

**MIC and MBC values determination:** The minimum inhibitory concentration (MIC) is the minimum amount of an extract that suppresses the growth of bacteria and the minimum

bactericidal concentration (MBC) is the minimum amount of an extract necessary for the killing of bacteria. Both find extensive uses as significant parameters of microbiology and tests for susceptibility against microbes.

The MIC and MBC values of this study were measured using the microdilution test combined with a colorimetric assay with iodinitrotetrazolium chloride (INT). The *C. mangga* (500 mg/mL) extracts were serially diluted in a 96-well plate with 100 µL of nutrient broth (NB). One hundred microliters of test bacterial suspension holding different amounts of *C. mangga* extract was added to each well. The 96-well plates were incubated overnight at 37°C. After incubation, 50 µL INT solution was added to each well and further incubated for 30 min at 37°C. The lowest concentration of *C. mangga* extract that suppressed the apparent growth of bacteria was defined as the MIC. The MBC is the lowest amount that eliminates the bacteria and indicates no color change after the addition of INT<sup>3,22-24</sup>.

**Data analysis:** The MIC and MBC values were analyzed using SPSS (version 28). The experimental design employed was a Completely Randomized Design (CRD) with three replicates

per treatment, each replicate forming three plates. Statistical differences among treatments were evaluated using One-way Analysis of Variance (ANOVA). Mean comparisons were then conducted using Duncan's Multiple Range Test (DMRT). Differences were considered statistically significant at a p-value of less than 0.05.

## RESULTS

**MIC and MBC values:** The antibacterial activity of *C. mangga* extracts varied depending on the solvent used and the bacterial strain assessed. The dichloromethane extract showed the strongest inhibitory effect, showing the lowest MIC value of 0.049 mg/mL against multidrug-resistant *K. pneumoniae* and *S. maltophilia*. Ethanol and hexane extracts also presented moderate activity, with MIC values ranging from 0.098-12.5 mg/mL across the tested strains. Notably, the ethanol extract was effective against *A. baumannii* (0.098 mg/mL), Colistin-resistant *P. aeruginosa* (0.39 mg/mL) and multidrug-resistant *K. pneumoniae* (0.39 mg/mL) (Table 2). In contrast, weaker activity (MIC<sub>≥</sub>12.5 mg/mL) was seen across all extracts against *K. oxytoca*, *S. aureus* and

Table 2: MIC values of *Curcuma mangga* extract against pathogenic bacteria

Bacterial strain	Average of MIC values* (mg/mL)		
	Ethanol extracts	Hexane extracts	Dichloromethane extracts
Multidrug-resistant <i>Klebsiella pneumoniae</i>	0.39 <sup>e</sup>	0.78 <sup>c</sup>	0.049 <sup>f</sup>
<i>Klebsiella pneumoniae</i> 101	0.78 <sup>d</sup>	1.56 <sup>b</sup>	3.125 <sup>b</sup>
<i>Klebsiella oxytoca</i> TISTR556	12.5 <sup>a</sup>	12.5 <sup>a</sup>	12.5 <sup>a</sup>
<i>Staphylococcus aureus</i> TISTR1466	6.25 <sup>b</sup>	12.5 <sup>a</sup>	12.5 <sup>a</sup>
<i>Staphylococcus epidermidis</i> TISTR518	6.25 <sup>b</sup>	12.5 <sup>a</sup>	12.5 <sup>a</sup>
<i>Klebsiella pneumoniae</i> TISTR1383	0.78 <sup>d</sup>	12.5 <sup>a</sup>	12.5 <sup>a</sup>
<i>Acinetobacter baumannii</i>	0.098 <sup>f</sup>	0.098 <sup>e</sup>	0.098 <sup>e</sup>
<i>Stenotrophomonas maltophilia</i>	3.125 <sup>c</sup>	0.049 <sup>f</sup>	0.78 <sup>c</sup>
<i>Proteus mirabilis</i>	-	-	12.5 <sup>a</sup>
Colistin-resistant <i>Pseudomonas aeruginosa</i>	0.39 <sup>e</sup>	0.39 <sup>d</sup>	0.39 <sup>d</sup>
p-value	<0.0001	<0.0001	<0.0001

\*Means (n = 3) in the column followed by the same common letter were not significantly different (DMRT, p>0.05)

Table 3: MBC values of *Curcuma mangga* extract against pathogenic bacteria

Bacterial strain	Average of MBC values* (mg/mL)		
	Ethanol extracts	Hexane extracts	Dichloromethane extracts
Multidrug-resistant <i>Klebsiella pneumoniae</i>	6.25 <sup>c</sup>	6.25 <sup>c</sup>	25 <sup>a</sup>
<i>Klebsiella pneumoniae</i> 101	12.5 <sup>b</sup>	12.5 <sup>b</sup>	25 <sup>a</sup>
<i>Klebsiella oxytoca</i> TISTR556	25 <sup>a</sup>	25 <sup>a</sup>	25 <sup>a</sup>
<i>Staphylococcus aureus</i> TISTR1466	12.5 <sup>b</sup>	25 <sup>a</sup>	25 <sup>a</sup>
<i>Staphylococcus epidermidis</i> TISTR518	25 <sup>a</sup>	25 <sup>a</sup>	25 <sup>a</sup>
<i>Klebsiella pneumoniae</i> TISTR1383	12.5 <sup>b</sup>	25 <sup>a</sup>	25 <sup>a</sup>
<i>Acinetobacter baumannii</i>	6.25 <sup>c</sup>	25 <sup>a</sup>	25 <sup>a</sup>
<i>Stenotrophomonas maltophilia</i>	6.25 <sup>c</sup>	3.125 <sup>d</sup>	1.56 <sup>c</sup>
<i>Proteus mirabilis</i>	-	-	25 <sup>a</sup>
Colistin-resistant <i>Pseudomonas aeruginosa</i>	6.25 <sup>c</sup>	12.5 <sup>b</sup>	12.5 <sup>b</sup>
p-value	<0.0001	<0.0001	<0.0001

\*Means (n = 3) in the column followed by the same common letter were not significantly different (DMRT, p>0.05)

*S. epidermidis*. No detectable inhibition was observed for *P. mirabilis* with ethanol and hexane extracts. These findings suggested that *C. mangga* extracts, particularly those derived using dichloromethane, possess promising antibacterial potential against certain resistant pathogens.

The bactericidal activity of *Curcuma mangga* extracts, as indicated by minimum bactericidal concentration (MBC) values, varied across bacterial strains and solvent types. The dichloromethane extract demonstrated the strongest bactericidal effect against *S. maltophilia*, with the lowest MBC value of 1.56 mg/mL. Ethanol and hexane extracts also showed moderate activity, with MBC values ranging from 3.125-25 mg/mL. The lowest MBCs were observed for multidrug-resistant *K. pneumoniae* (6.25 mg/mL for ethanol and hexane), *A. baumannii* (6.25 mg/mL for ethanol) and Colistin-resistant *P. aeruginosa* (6.25-12.5 mg/mL across all extracts) (Table 3). However, all extracts exhibited relatively weak bactericidal activity (MBC = 25 mg/mL) against *K. oxytoca*, *S. aureus* and *S. epidermidis*. No bactericidal effect was detected for *P. mirabilis* in ethanol and hexane extracts. These results suggest that *C. mangga* extracts possess selective bactericidal properties, with the dichloromethane extract being particularly effective against certain resistant strains.

## DISCUSSION

The antibacterial properties of *C. mangga* extracts have been demonstrated in their activity against *Aeromonas hydrophila*, a pathogenic bacterium of concern in aquaculture and clinical settings. *In vitro* assays revealed that the MIC of the extracts was determined to be 6.25 mg/mL, indicating the lowest concentration at which visible bacterial growth was inhibited. Furthermore, the MBC was established at 50 mg/mL, representing the lowest concentration required to achieve complete bacterial eradication<sup>25</sup>. *C. mangga* oil exhibited the most potent and broad-spectrum antimicrobial activity. It effectively inhibited the growth of all evaluated microorganisms, including *B. cereus*, *S. aureus*, *Candida albicans* and *Cryptococcus neoformans*. The observed MIC and minimum bactericidal/fungicidal concentration (MBC/MFC) values ranged from 0.1 to 11.1 µL/mL underscoring its significant potential as a natural antimicrobial agent<sup>16</sup>.

The ethanol extract of *C. mangga* rhizome demonstrated superior antibacterial activity compared to chloroform and n-hexane extracts. Specifically, the ethanol extract exhibited an MIC of 3.13% and an MBC of 6.25% against *S. aureus*. In the case of *E. coli*, the MIC and MBC values were determined to be 6.25 and 12.50%, respectively<sup>17</sup>. The MIC value (0.39 mg/mL)

and MBC values (ranging from 6.25-12.5 mg/mL) observed in this study were notably higher than those previously reported for *C. domestica*, *C. xanthorrhiza* and *C. mangga* against multidrug-resistant *P. aeruginosa*, which were 125 µg/mL, 250 µg/mL and 125 µg/mL, respectively. This discrepancy may reflect differences in extract composition, bacterial strain susceptibility, or methodological variations and warrants further investigation to elucidate the underlying factors<sup>26</sup>. This study demonstrated that the MIC values for multidrug-resistant *K. pneumoniae* (ranging from 0.049-0.78 mg/mL), as well as for *K. pneumoniae* 101 and *K. pneumoniae* TISTR1383 (both at 0.78 mg/mL), were markedly lower than those reported in prior literature. Specifically, previous studies have shown that the ethanolic extract of *C. xanthorrhiza* exhibited antibacterial activity against *K. pneumoniae* with a MIC value of 1.25 mg/mL. These comparative results suggest that the extracts investigated in the current study may offer superior antibacterial efficacy, particularly against resistant *K. pneumoniae* strains<sup>27</sup>.

The findings of this study revealed that the MIC and MBC values of the tested extract against *P. mirabilis* were 12.5 and 25 mg/mL, respectively. These values are notably higher than those reported in previous studies, such as the work on the ethanolic extract of *Curcuma longa* L. rhizomes, which demonstrated superior antibacterial efficacy with MIC and MBC values of 0.048 and 0.39 mg/mL, respectively, against *P. mirabilis*. While previous studies reported no observable antibacterial activity of the extract against *A. baumannii*, the findings of the present study demonstrate that the *C. mangga* extract exhibited both growth-inhibitory and bactericidal effects against this pathogen. This notable discrepancy suggests that *C. mangga* may possess previously unrecognized antimicrobial potential against *A. baumannii*, warranting further investigation into its bioactive constituents and mechanisms of action<sup>19</sup>.

To date, there have been no documented reports on the antibacterial activity of *C. mangga* against antibiotic-resistant bacterial strains, including multidrug-resistant *K. pneumoniae*, *A. baumannii*, *S. maltophilia*, *P. mirabilis* and colistin-resistant *P. aeruginosa*. This gap in the literature highlights the need for further investigation into the potential of *C. mangga* as a source of novel antimicrobial agents targeting resistant pathogens.

## CONCLUSION

The first findings of this study reveal that *C. mangga* extracts, particularly the dichloromethane fraction, exhibited the most pronounced antibacterial and

bactericidal activities. This extract achieved the lowest MIC and MBC values against multidrug-resistant strains of *K. pneumoniae* and *S. maltophilia*. In addition, the ethanol and hexane extracts demonstrated moderate antimicrobial efficacy, notably against *A. baumannii* and colistin-resistant *P. aeruginosa*. These results underscore the potential of *C. mangga* as a valuable natural source for the development of plant-based antimicrobial agents targeting antibiotic-resistant pathogens.

### SIGNIFICANCE STATEMENT

This study provides the first comprehensive evidence of the antibacterial and bactericidal potential of *C. mangga* extracts against both antibiotic-resistant and reference pathogenic bacterial strains. The pronounced efficacy of the dichloromethane extract, particularly against multidrug-resistant *K. pneumoniae* and *S. maltophilia*, highlights its potential as a natural antimicrobial agent. These findings contribute to the growing body of research on plant-based alternatives for addressing the global challenge of antibiotic resistance and support the future development of *C. mangga*-derived therapeutic products.

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