Full Paper
Journal of Nano Materials Impact 2025, 01 (02) 17-23

Antibacterial Activity of Nutmeg (*Myristica fragrans*) Seeds Extracts Against Pathogenic Bacteria Isolated from Infected Wounds

Ameera Alan Jasim[1], Fitua Al-Saedi*[,2,3], Basant Salah Mahmoud[4]

[1]Department of Clinical Laboratory Sciences, College of Pharmacy, Mustansiriyah University, Baghdad, IRAQ. [2]Department of Clinical Laboratory Sciences, College of Pharmacy, Mustansiriyah University, Baghdad, IRAQ. [3]Polymers research unit, College of Science, Mustansiriyah University, Baghdad, IRAQ, [4]Hormones Department, Institute of Medical Research and Clinical Studies, National Research Centre, Giza, Dokki, EGYPT.

Abstract

One of the most common hospital-acquired infections is a wound infection. Bacterial infection of wounds is a critical problem, and the healing of wound infections remains an important concern for surgeons. The problem has been exaggerated because of the uncontrolled and quickly spreading resistance to the existing antimicrobial agents. To overcome bacterial resistance to antibiotics, a variety of alternatives have been suggested. Medicinal plants extracted through various methods, as well as their nano-formulations, have emerged as promising sources for novel antimicrobial agents**.** This study aimed to evaluate the antibacterial efficacy of *Myristica fragrans* (Nutmeg) seed extracts against multidrug-resistant pathogenic bacteria isolated from infected wounds**.** A total of 50 wound swab samples were collected from patients who were admitted to hospitals in Baghdad. Different biochemical tests were used to identify the isolated bacteria. An antibiotic susceptibility test was also done. *M. fragrans* seeds' ethanolic and aqueous crude extracts were prepared. The antibacterial activity of the extracts was investigated using the agar well diffusion method**.** Eight genera of bacterial agents were isolated and identified: *Escherichia coli* (58.3%), *Acinetobacter baumannii* (4.2%), *Klebsiella pneumoniae* (8.3%), *Pseudomonas aeruginosa* (20.8%), *Enterobacter* (8.3%), *Staphylococcus aureus* (80%), *Staphylococcus epidermidis* (11.5%), and *Streptococcus sp.* (7.7%). Antibiotic susceptibility test showed that the isolated bacteria were multidrug-resistant. The results revealed the potential activity of *M. fragrance* seeds extracts against the isolated bacteria. Hot ethanolic extract showed antibacterial activity in all concentrations (2 mg/ml, 4 mg/ml, and 8 mg/ml). The highest inhibition zone was 17.00±2.000, 14.67±0.77, and 14.33±1.5 mm at a concentration of 8 mg/ml against *A. baumannii*, *P. aeruginosa*, and *E. coli*, respectively. Followed by *K. pneumoniae*, *Enterobacter sp.*, and *S. aureus*, the zone of inhibition was 13.67±1.528 mm and 12.67±1155 mm, respectively. Hot aqueous extract also showed activity. The highest inhibition zone at a concentration of 8 mg/ml was against *Enterobacter sp.* (15.00±0.000 mm), followed by *A. baumannii* (14.67±0.577mm), *K. pneumoniae* (14.33±0.577 mm), *P. aeruginosa* (13.67±0.577 mm), *E. coli* (13.00±1.000 mm), and *S. aureus* (12.00±0.000mm). *M. fragrans* seed extracts showed antibacterial activity against both gram-positive and gram-negative multidrug-resistant pathogenic bacteria. Based on this work findings, *M. fragrans* extracts have a broad spectrum of antibacterial activity.

Keyword: wound infection • pathogenic bacteria • antibiotics • Myristica fragrans • extracts.

Introduction

One of the most popular hospital-acquired infections is wound infection[1]. Infection of wounds occurs by invasion and proliferation by pathogens that are able to trigger host response[2-4]. The existence of pathogens within the wound leads to tissue damage and delays wound healing[5]. A diversity of microorganisms can infect wounds, such as bacteria, fungi, and parasites.

***Address for correspondence:** Dr. Fitua Al-Saedi, Department of Clinical Laboratory Sciences, College of Pharmacy, Mustansiriyah University, Baghdad, Iraq.

E-mail: fituaaziz@uomustansiriyah.edu.iq

ORCID: https://orcid.org/0000-0003-0734-8192

Info:
Submitted: 19 March 2025; **Revised:** 03 May 2025;
Accepted: 02 Aug. 2025; **Published:** 18 Aug. 2025.
https://doi.org/10.71109/nmi.2025.1.2.13

Various studies on septic wounds that have been done showed the commonly isolated bacteria were *P. aeruginosa*, *S. aureus*, *Klebsiella sp.*, *E. coli*, *Proteus spp.*, and *Acinetobacter sp.*[6]. In addition, *Salmonella sp.*, which is known to cause gastrointestinal infections, can cause skin and soft tissue infections in immunocompromised patients[7]. The control of wound infections has become challenging due to the widespread bacterial resistance to antibiotics and the greater incidence of infections caused by poly microbial flora[8]. To overcome bacterial resistance to antibiotics, a variety of antibiotic alternatives have been suggested, such as bacteriophages, vaccines, antimicrobial peptides, medicinal plant extracts, etc.[9]. Medicinal Plants act as a valuable source of active composites that have antimicrobial activity[10-12]. They contain a variety of active compounds such as flavonoids, polyphenols, alkaloids, tannins, and saponins that have antioxidant and antimicrobial activity[13, 14]. *Myristica fragrans*, generally known as nutmeg, has different components that belong to terpenoids, lignans, flavonoids, steroids, and saponins[15-18]. These compounds have biological activities such as antioxidant[16], anti-inflammatory, and antimicrobial[19]. Nutmeg seed extracts and their nano-formulations offer a promising solution to the growing challenge of antibiotic resistance and microbial

infections. Several studies have demonstrated that extracts of *M. fragrans* seeds inhibit the growth of both gram-positive and Gram-negative bacteria, including *Staphylococcus aureus*, *Escherichia coli*[20]. The mechanism is believed to involve disruption of bacterial cell membranes and inhibition of essential bacterial enzymes, making *M. fragrans* a promising source of natural antimicrobials[21]. Nanoparticle formulations of *Myristica fragrans* seed extracts have shown antibacterial activity due to improved bioavailability, surface area, and cellular uptake**.** It has been shown that *M. fragrans*-based Ag nanoparticles have potent antibacterial activity against pathogens including *Klebsiella pneumoniae*, *Bacillus subtilis*, and *Salmonella typhi* [22]. The nanoparticles disrupt bacterial cell walls, generate reactive oxygen species (ROS), and interfere with DNA replication. The current study aims to investigate the antibacterial efficacy of *Myristica fragrans* (Nutmeg) seeds extracts against different multidrug resistant pathogenic bacteria isolated from infected wounds.

Experimental Part

Methods

Isolation and Identification of Pathogenic Bacteria

A Total of 50 wound swab samples were collected from patients who were admitted to Baghdad Hospitals. After adequately washing the wound with sterile distilled water in order to avoid contamination, one swab from each patient was obtained. Within an hour of sampling, the samples were transferred to a microbiology laboratory. Swabs were immediately inoculated on MacConkey agar, Chocolate agar, Blood agar, Mannitol salt agar, and Cetrimide agar at 37 °C for 24 to 48 hours. Bacterial identification was done using biochemical tests: Motility, Kligler Iron, Coagulase, Catalase, Urease, Citrate, and Indole.

Antibiotic Susceptibility Test

The sensitivity to the antibiotics was determined by the Kirby-Bauer disc diffusion method and described according to the guidelines of the Clinical Laboratory Standards National Committee (2020). Muller-Hinton (MH) was used as agar media. The density of cultured bacteria was compared with a 0.5 McFarland standard that provides an optical density comparable to the density of a bacterial suspension with a 1.5×10^8 colony-forming units (cfu/ml). A small amount of bacterial dilution is taken with a cotton swab and spread on the plates. Antibiotic discs were placed on the inoculated plates. Incubation of plates was done at 37°C for 24 h, and the zone of inhibition around the disks was measured using a ruler. Three replicates were prepared for each microorganism[23].

The antibiotic discs used for gram-positive bacteria were Vancomycin (30 mg/disc), Tetracycline (30 mg/disc), fusidic acid (20 mg/disc), Clindamycin (2 mg/disc), Tobramycin (10 mg/disc), Erythromycin (15 mg/disc) Teicoplanin (30 mg/disc, levofloxacin (5 mg/disc) Netilmicin (30 mg/disc), Cefoxitin (30 mg/disc). The antibiotic discs used for gram-negative bacteria were used Amikacin (30 mg/disc), Ciprofloxacin (5 mg/disc), Gentamycin (10 mg/disc), Meropenem (10 mg/disc), Trimethoprim (5mg/disc), Azithromycin (15 mg), Ceftazidime (30 mg/disc), Colistin (5 mg/disc), Cefotaxime (30mg/disc), Amoxicillin (30 mg/disc).

Collection of Plants

The dried *Myristica fragrans* (Nutmeg) seeds were purchased from the local market.

Preparation of the Ethanolic Extract

The seed powder (50 g) was extracted with 350 ml of 70% ethanol for 8 hrs. using Soxhlet equipment. The extract was filtered, and the filtrate was then evaporated using a rotary evaporator[24]. The dried extract was kept at 4˚C until evaluation of its antibacterial activities.

Preparation of the Aqueous Extraction

The powdered samples (40g) were soaked in 100 ml of water for 48 hours. Then, it was filtered using a muslin cloth; the filtrate was re-filtered using a Whitman filter paper[25].

Antibacterial assay

The anti-bacterial activity is assessed using the agar well diffusion method[26]. Plant extracts were diluted in sterile DMSO to prepare the following concentrations: 8 mg/ml, 4 mg/ml, and 2 mg/ml. Muller-Hinton agar was inoculated with one of the isolated bacteria (10^8 cfu /ml, adjusted to 0.5 McFarland standard). 100µl from each concentration was added to 6mm diameter wells. Amoxicillin/Clavulanic acid (30 µg/disc) was used as a positive control. Plates are incubated at 37 ºC for 24 hours. All the experiments were conducted in triplicate.

Statistical Analysis

The data from the study in MS Excel and analyzed using SPSS software version 24. The data were expressed as Mean ± Standard deviation. A one-way analysis of variance was carried out at $P < 0.05$, and the Waller-Duncan Test was used to ascertain the source of the variation among the isolates.

Results and Discussion

Results

Identification of Pathogenic Bacteria Isolated from Wound Infection

On the Blood agar, MacConkey agar, chocolate agar, Mannitol salt agar, and Cetrimide agar, the colonies were isolated and identified. Different biochemical tests were used to confirm the identified pathogenic bacteria, as shown in **Table 1**.

Distribution of Bacterial Isolates from the Infected Wound Samples

Eight genera of bacterial agents were isolated and identified (See **Table 2**). Five genera belong to gram-negative bacteria, including *E. coli* (58.33%), *Acinetobacter baumannii* (4.17%), *Klebsiella pneumoniae* (8.33%), *Pseudomonas aeruginosa* (20.83%), and *Enterobacter sp.* (8. 33%). In addition, three genera belong to gram-positive bacteria. Those include: *Staphylococcus aureus* (80.77%), *S. epidermidis* (11.5%), and *Streptococcus* sp. (7.69%).

Table 1. Biochemical tests are used to identify the pathogenic bacteria isolated from the infected wounds.

Bacteria	Motility	Kligler Iron	Coagulase	Catalase	Urease	Citrate	Indole
E. coli	P	P	N	N	N	N	P
P. aeruginosa	p	N	N	P	N	P	N
A. baumanii	N	N	N	N	N	P	N
Enterococcus sp.	P	P	N	N	N	P	N
S. aureus	N	N	p	P	N	N	N
S. epidermidis	N	N	N	P	P	N	N
Streptococcus sp.	N	N	N	N	P	N	N
K. pneumonia	N	P	N	P	P	P	N

P=positive, N=negative

Table 2. Distribution of bacterial isolates from the infected wound samples.

Isolates	Number of isolates	Frequency (%)
S. epidermidis	3	11.54
S. aureus	21	80.77
Streptococcus sp.	2	7.69
A. baumannii	1	4.17
E. coli	14	58.33
Enterobacter sp	2	8.33
K. pneumonia	2	8.33
P. aeruginosa	5	20.83
Total	50	100%
Isolates	**Number of isolates**	**Frequency %**

Antibiotic Susceptibility Patterns of Gram-Positive Isolates

In the current study, antibiotic susceptibility patterns of the gram-negative isolates showed that Amikacin (67.7%) and Amoxicillin (64.9%) were the most effective antibiotics (See **Figure 1** and **Table 3**).

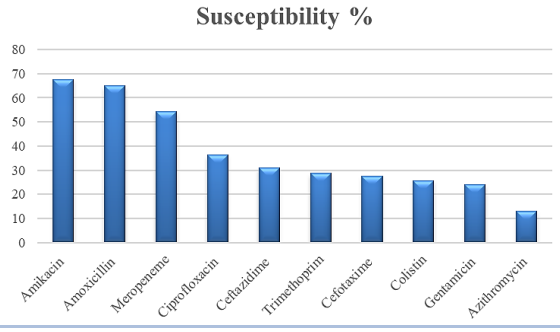

Figure 1. Susceptibility percentage of all gram-negative pathogens to antimicrobial agents.

Table 3. Antibiotic susceptibility patterns of gram-positive pathogens isolated from infected wounds.

Antibiotics	Bacteria					
	S. epidermidis N=3		*S. aureus* N=21		*Streptococcus* sp N=2	
	S%	R%	S%	S%	R%	S%
Teicoplanin	66.7	33.3	66.7	33.3	50	50
Vancomycin	33.3	66.7	42.9	57.1	0	100
Tetracycline	66.7	33.3	28.6	71.4	50	50
Fusidic acid	100	0	47.6	52.4	100	0
Levofloxacin	66.7	33.3	47.6	52.4	50	50
Clindamycin	0	100	52.4	47.6	100	0
Tobramycin	33.3	66.7	38.1	61.9	0	100
Netilmicin	66.7	33.3	61.9	38.1	50	50
Erythromycin	100	0	66.7	33.3	100	0
Cefoxitin	66.7	33.3	52.4	47.6	0	100

Tobramycin (76.2%) followed by Vancomycin (74.6%) were the most effective antibiotics against gram-positive pathogenic isolates (See **Figure 2** and **Table 4**).

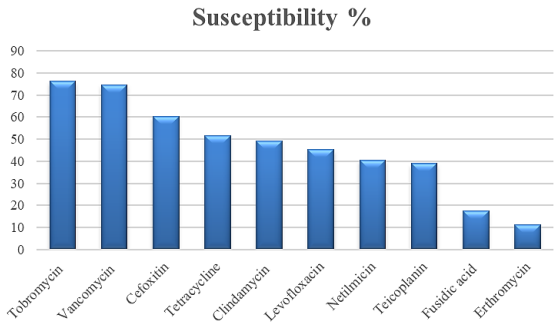

Figure 2. Susceptibility percentage of all gram-positive pathogens to antimicrobial agents.

Table 4. Antibiotic susceptibility patterns of gram-positive pathogens isolated from infected wounds.

Antibiotics	Bacteria					
	A. baumannii N=1		*E. coli* N=14		*Enterobacter* sp. N=2	
	R%	S%	R%	S%	R%	R%
Amikacin	100	0	21.4	78.6	0	100
Ciprofloxacin	100	0	21.4	78.6	50	100
Gentamicin	100	0	50	50	100	100
Meropeneme	100	0	28.6	71.4	50	100
Trimethoprim	100	0	57.1	42.9	0	100
Azithromycin	100	0	85.7	14.3	50	100
Ceftazidime	100	0	85.7	14.3	0	100
Colistin	0	100	71.4	28.6	100	0
Cefotaxime	100	0	42.9	57.1	100	100
Amoxicillin	0	100	85.7	14.3	0	0

Evaluation of Antibacterial Activity of Myristica Fragrance Extracts

The results shown in **Table 5** and **Figure 3** revealed the potential activity of *M. fragrance* hot ethanolic and hot aqueous extracts against different bacteria isolated from wound infection.

Hot ethanolic extract showed antibacterial activity of all the tested concentrations (2 mg/ml, 4 mg/ml, and 8 mg/ml). The highest inhibition zone was 17.00±2.000, 14.67±0.77, and 14.33±1.5 mm at a concentration of 8 mg/ml against *A. baumannii*, *Pseudomonas aeruginosa*, and *E. coli*, respectively. Followed by *K. pneumoniae*, the zone of inhibition was 13.67±1.528 mm. Hot ethanolic extract also showed inhibitory effect against *S. aureus.* The highest inhibition zone against *S. aureus* at a concentration of 8 mg/ml was 12.67±1. 155 mm.

Hot aqueous extract (**Table 5** and **Figure 4**) shows antibacterial activity at all the used concentrations (2 mg/ml, 4 mg/ml, and 8 mg/ml). The highest inhibition zone at a concentration of 8 mg/ml was against *Enterobacter sp.* (15.00±0.000 mm), followed by *A. baumannii* (14.67±0.577 mm), *K. pneumoniae* (14.33±0.577 mm), *P. aeruginosa* (13.67±0.577 mm), and *E. coli* (13.00±1.000 mm). In addition, hot aqueous extract inhibits *S. aureus* growth at all concentrations; the highest inhibition zone at the concentration of 8 mg/ml was (12.00±0.000 mm).

Table 5. Antibiotic susceptibility patterns of gram-positive pathogens isolated from infected wounds.

Extracts	Pathogenic bacteria	Mean(mm)+SD			Amoxicillin -clavulanic 30 (µg/disc)
		2 (mg/ml)	2 (mg/ml)	2 (mg/ml)	
Hot Ethanolic	***A. baumannii***	14.00±3.464c	16.00±1.000c	17.00± 2.000c	R
	K. pneumoniae	13.00±1.732ab	13.67±1.528bc	13.67±1.528abc	13 mm
	E. coli	12.67±1.155bc	12.33±2.082bc	14.33±1.528abc	16 mm
	Enterobacter sp.	12.33±0.577abc	12.67±1.155bc	13.67±1.528 a	14 mm
	S. aureus	10.67±1.155b	12.00±1.732bc	12.67±1.155ab	14 mm
	P. aeruginosa	13.00±0.000a	14.33±0.577b	14.67±0.77bc	16 mm
Hot aqueous	***A. baumannii***	14.00±0.000a	14.67±0.577b	14.67+0.577bc	R
	K. pneumoniae	13.33+0.577abc	13.67+0.577b	14.33+0.577bc	13mm
	E. coli	12.33+0.577ab	12.67+0.577b	13.00+1.000a	16 mm
	Enterobacter sp.	12.00+0.000a	14.67+0.577b	15.00+0.000a	14mm
	S. aureus	10.33±0.577ab	11.00± 1.000a	12.00± 0.000a	14 mm
	P. aeruginosa	12.33+0.577ab	13.00+0.000a	13.67+0.577b	16 mm

Different letters along the column indicate significant variation (P < 0.05) according to Waller Duncan statistics.

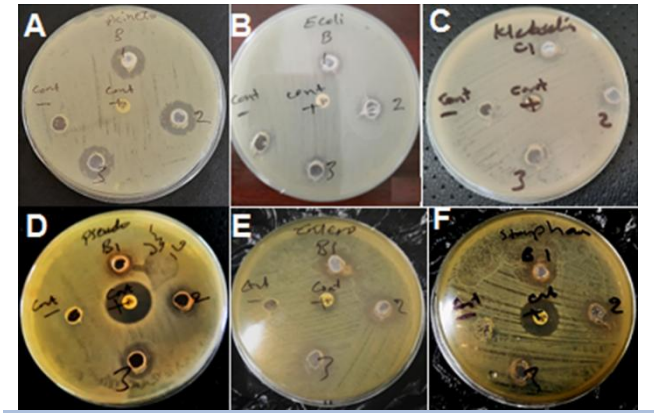

Figure 3. Agar plates showing the zone of inhibition around wells for hot ethanolic extract against pathogenic bacteria related to wound infection. (A) *A. baumannii*, (B) *E. coli*, (C) *K. pneumonia*, (D) *P. aeruginosa*, (E) *Enterobacter sp.*, and (F) *S. aureus*.

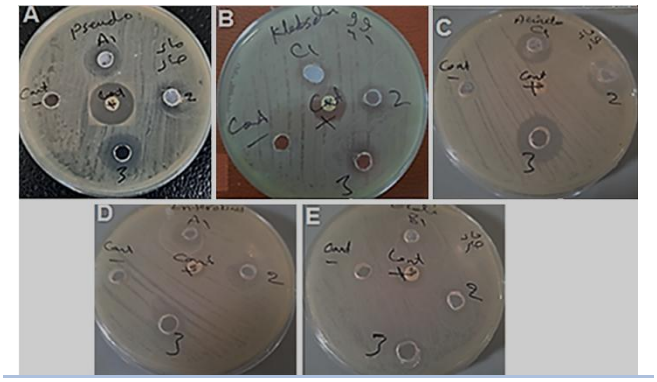

Figure 4. Agar plates showing the zone of inhibition around wells for hot aqueous extract against pathogenic bacteria related to wound infection. (A) *P. aeruginosa*, (B) *A. baumannii*, (C) *K. pneumonia*, (D) *Enterobacter sp.*, and (E) *E. coli*.

Discussion

Herein, *S. aureus* was isolated in the highest percentage (80.77 %). *S. aureus* is known to cause wound infection, both Nosocomial infection (hospital-acquired) and community-acquired infection[27]. A previous study showed that out of 50 wound swabs, *S. aureus* was isolated at a percentage of 68% [27]. Another study done in Iraq showed that *S. aureus* was isolated in 34% of all cases[28]. Another study, which is in the same line, revealed that *S. aureus*, followed by *E. coli*, Beta-hemolytic Streptococci, *Klebsiella aerogenes*, *Pseudomonas aeruginosa*, *Proteus spp., and Streptococcus faecalis* were isolated from the infected wounds[29]. On the other hand, the current study found that *E coli* was isolated at the highest percentage among gram-negative bacteria (58.33%), followed by *P. aeruginosa* (20.83%), *Klebsiella pneumonia* and *Enterobacter* sp. (8.33%), and *Acinetobacter baumannii* (4.17%). The highest percentage of the isolated *E coli* among other gram-negative bacteria could be because of the contamination in place of surgery. These findings are in agreement with another study's results in Patna Medical College and Hospital in India, which proved the maximum rate of *E. coli* (27.7%)[30].

Antibiotic susceptibility results of gram-positive bacteria (Table 3) showed that *S. aureus* was highly resistant to Teicoplanin and erythromycin at a percentage of 66.7%. *S. epidermidis* showed the highest resistance to Fusidic acid, erythromycin, and clindamycin at a percentage of 100%. A previous study showed that the percentage of resistance of *S. aureus* isolates to erythromycin was 45.6%, fusidic acid (44.0%), and clindamycin (42.3%). None of the *S. aureus* isolates exhibited resistance to teicoplanin[31].

Another study showed that among fusidic acid-resistant *S. epidermidis* isolates, 80.0% were resistant to erythromycin, and 65.5% were resistant to clindamycin [32].

This study showed alarming results about the antibiotic susceptibility test of gram-negative bacteria (Table 4), which revealed that *A. baumannii* was highly resistant to Amikacin, Ciprofloxacin, Gentamicin, Meropenem, Trimethoprim, Azithromycin, Ceftazidime, and Cefotaxime at a rate of 100%. Enterobacter was highly resistant to Gentamicin**,** Colistin, and Cefotaxime at a percentage of 100%. *Klebsiella pneumoniae* showed the highest resistance to Trimethoprim, Azithromycin, Ceftazidime, Colistin, and Cefotaxime at a percentage of 100%. *Pseudomonas aeruginosa* was highly resistant to Trimethoprim, Azithromycin, and colistin (100%). *E. coli* was highly resistant to Azithromycin, Ceftazidime, and amoxicillin at a percentage of 85.7%.

Recently, it has been reported that 67% of the *A. baumannii* isolates were multidrug-resistant strains. They exhibited high resistance to most of the tested antibiotics; the highest was to ceftriaxone (83%) and ceftazidime (75%)[33]. Another study

showed resistance in more than 80% of *A. baumannii* isolates to the carbapenems[34]. And 99% were carbapenem-resistant isolates[35]. In a previous study, the results revealed that *K. pneumoniae* isolates displayed a high percentage of resistance to most of the tested antibiotics[36]. It has been shown that *K. pneumoniae* the rate of resistance to cefotaxime and ceftriaxone (97%), trimethoprim-sulfamethoxazole (93.9%), amikacin (93.2%), and imipenem (81.1%). Another study conducted in Iraq, the results pointed to a high resistance rate of *P. aeruginosa* to amoxicillin, gentamicin, trimethoprim, and amikacin[37]. Multi-drug resistant (MDR) pathogens are resistant to several classes of antimicrobial agents. MDR is considered one of the most essential problems faced by the healthcare sector at the moment, and the discovery of compounds active against these pathogens is of current importance.

To overcome bacterial resistance to antibiotics, a variety of antimicrobial alternatives have been suggested. Medicinal plants act as a valuable source of active composites that have antimicrobial activity[11-13]. *Myristica fragrant* is recognized to show strong antimicrobial activity against different pathogens due to its contents of active components[21, 22]. Studying the antimicrobial activity of the seeds of *Myristica fragrans* extract against MDR pathogens is worth noting. In the present study, antibacterial activity of Nutmeg seed extracts was tested against different pathogenic bacteria that cause wound infection, including *Enterobacter sp.*, *A. baumannii*, *K. pneumoniae*, *P. aeruginosa*, *E. coli*, and *S. aureus*. The results showed that the seed extracts have antibacterial activity against all tested bacteria, including the MDR bacteria. The major components of *Myristica fragrans* are alkyl benzene derivatives (myristicin, elemicin, safrole, etc.), terpenes, alpha-pinene, beta-pinene, myristic acid, trimyristin, neolignane (myrislignan), and macelignan[38-41]. It has been reported that trimyristin, an active compound obtained from the seed of *Myristica fragrans*, has antibacterial activity against both gram-positive and gram-negative bacteria[42]. Another study's results revealed that nutmeg oil, including sabinene, 4-terpineol, myristicin, and alpha-pinene, has antibacterial activity against *E. coli*, *S. aureus*, and *P. aeruginosa*[43]. Moreover, earlier results demonstrated that ethanol extracts of Nutmeg inhibit the growth of *Streptococcus mutans*, *Streptococcus mitis*, *Streptococcus salivarius*, *Aggregatibacter*, *actinomycetemcomitans*, and *Porphyromonas*[44]. The antimicrobial activity of the seed oil of Nutmeg against clinical isolates of *E. coli*, *K. pneumoniae*, *P. mirabilis*, *P. vulgaris*, *P. aeruginosa, and S. aureus* has been reported previously. Previous results showed that the minimum inhibitory concentration (MIC) of oil of seed was 1 mg/mL for *E. coli*, *P. mirabilis*, and *P. aeruginosa*, and > 1 mg/mL for *P. vulgaris* and *K. pneumoniae*[21]. Another study's results showed the antimicrobial activity of the seed oil of Nutmeg against MRSA, *E. cloacae*, and *Proteus sp.*[45]. In literature, it has been shown that the mechanism of action of nutmeg seed extracts involves by disruption of bacterial cell membranes and inhibition of essential bacterial enzymes, making *M. fragrans* a promising source of natural antimicrobials[21].

Conclusion

The results of the current work demonstrate that Nutmeg (*Myristica fragrans*) extracts have antibacterial activity against multidrug-resistant pathogenic bacteria isolated from infected wounds, including *Enterobacter sp.*, *A. baumanii*, *K. pneumoniae*, *P. aeruginosa*, *E. coli*, and *S. aureus*. Based on these findings, *Myristica fragrans* extracts have a broad spectrum of antibacterial activity, and they could be a potential source of new antibacterial agents. Further studies, *in vivo*, are recommended to determine the efficacy of these extracts in the treatment of bacterial infections.

Acknowledgements

The authors would like to thank Mustansiriyah University, Baghdad, Iraq, for its support in the present work.

Declaration of Competing Interests

All authors have made substantial contributions to the work and approved it for publication.

References

[1] J. M. Torpy, A. Burke, and R. M. Glass, "Wound Infections," *JAMA*, vol. 294, no. 16, p. 2122-2122, 2005.
https://doi.org/10.1001/jama.294.16.2122.

[2] W. A. Muller, "How Endothelial Cells Regulate Transmigration of Leukocytes in the Inflammatory Response," *The American Journal of Pathology*, vol. 184, no. 4, p. 886-896, 2014.
https://doi.org/10.1016/j.ajpath.2013.12.033.

[3] D. Vestweber, "How leukocytes cross the vascular endothelium," *Nature Reviews Immunology*, vol. 15, no. 11, p. 692-704, 2015.
https://doi.org/10.1038/nri3908.

[4] Y. Liu *et al.*, "Neutrophil heterogeneity and aging: implications for COVID-19 and wound healing," (in English), *Frontiers in Immunology*, Review vol. Volume 14 - 2023, 2023.
https://doi.org/10.3389/fimmu.2023.1201651.

[5] S. Omara *et al.*, "Predictors of Post-Caesarean Surgical Site Infections at Mubende Regional Referral Hospital, Central Uganda: Prospective Cohort Study (July–September 2023)," *International Journal of Women's Health*, p. 1939-1945, 2024.
https://doi.org/10.2147/IJWH.S482864.

[6] V. Puca *et al.*, "Microbial Species Isolated from Infected Wounds and Antimicrobial Resistance Analysis: Data Emerging from a Three-Years Retrospective Study," *Antibiotics*, vol. 10, no. 10,
https://doi.org/10.3390/antibiotics10101162

[7] C. T. Lin *et al.*, "Inferior Gluteal Artery Perforator Flap for Sacral Pressure Ulcer Reconstruction: A Retrospective Case Study of 11 Patients," (in eng), *Ostomy Wound Manage*, vol. 62, no. 1, p. 34-9, 2016.
PMID: 26779702.

[8] A. Ly, J. Henderson, A. Lu, E. Culham Doreen, and M. Wood Janet, "Osmoregulatory Systems of Escherichia coli: Identification of Betaine-Carnitine-Choline Transporter Family Member BetU and Distributions of betU and trkG among Pathogenic and NonpathogenicIsolates," *Journal of Bacteriology*, vol. 186, no. 2, p. 296-306, 2004.

https://doi.org/10.1128/JB.186.2.296-306.2004.

[9] M. J. Alwan, I. J. Lafta, and A. M. Hamzah, "Bacterial isolation from burn wound infections and studying their antimicrobial susceptibility," (in en), *Kufa Journal for Veterinary Medical Sciences*, vol. 2, no. 1, p. 121-131, 2011.

https://doi.org/10.36326/kjvs/2011/v2i14025.

[10] A. G. Rosyada *et al.*, "Aktivitas antibiofilm ekstrak etanol kulit bawang merah (Allium cepa L.) dalam menghambat pembentukan biofilm Staphylococcus aureus ATCC 25923," *Jurnal Kedokteran Gigi Universitas Padjadjaran*, vol. 35, no. 1, p. 33-40, 2023.

https://doi.org/10.24198/jkg.v35i1.42451.

[11] P. Jain, T. Satapathy, and R. K. Pandey, "Acaricidal activity and biochemical analysis of citrus limetta seed oil for controlling ixodid tick rhipicephalus microplus infesting cattle," *Systematic and Applied Acarology*, vol. 26, no. 7, p. 1350-1360, 2021.

https://doi.org/10.11158/saa.26.7.13.

[12] G. Casillas-Vargas *et al.*, "Antibacterial fatty acids: An update of possible mechanisms of action and implications in the development of the next-generation of antibacterial agents," *Progress in Lipid Research*, vol. 82, p. 101093, 2021.

https://doi.org/10.1016/j.plipres.2021.101093.

[13] S. MOHAPATRA, R. MOHANDAS, and R. P. KUMAR, "Comparison of the Anti-microbial Efficacy of Clove, Cinnamon, Turmeric, Nutmeg, and Peppermint Essential Oil against Oral Pathogens: An In-vitro Study," *Journal of Clinical & Diagnostic Research*, vol. 17, no. 5, 2023.

https://doi.org/10.7860/JCDR/2023/61363.17800.

[14] W. H. Al-Qahtani *et al.*, "Phyto-chemical and biological activity of Myristica fragrans, an ayurvedic medicinal plant in Southern India and its ingredient analysis," *Saudi Journal of Biological Sciences*, vol. 29, no. 5, p. 3815-3821, 2022.

https://doi.org/10.1016/j.sjbs.2022.02.043.

[15] S. K. Francis, B. James, S. Varughese, and M. S. Nair, "Phytochemical investigation on Myristica fragrans stem bark," *Natural Product Research*, vol. 33, no. 8, p. 1204-1208, 2019.

https://doi.org/10.1080/14786419.2018.1457670.

[16] J. Dzotam Kamgue *et al.*, "Further antibacterial compounds from Myristica fragrans," *Investigational Medicinal Chemistry and Pharmacology*, vol. 1, p. 5, 2018.

https://doi.org/10.31183/imcp.2018.00015.

[17] T. Oo *et al.*, "Effectiveness of co-cultured Myristica fragrans Houtt. seed extracts with commensal Staphylococcus epidermidis and its metabolites in antimicrobial activity and biofilm formation of skin pathogenic bacteria," *BMC Complementary Medicine and Therapies*, vol. 24, no. 1, p. 380, 2024.

https://doi.org/10.1186/s12906-024-04675-z.

[18] M. T. Ha, N. K. Vu, T. H. Tran, J. A. Kim, M. H. Woo, and B. S. Min, "Phytochemical and pharmacological properties of Myristica fragrans Houtt.: an updated review," *Archives of Pharmacal Research*, vol. 43, no. 11, p. 1067-1092, 2020.

https://doi.org/10.1007/s12272-020-01285-4.

[19] N. Suthisamphat *et al.*, "Anti-Helicobacter pylori, Anti-Inflammatory, Cytotoxic, and Antioxidant Activities of Mace Extracts from Myristica fragrans," *Evidence-Based Complementary and Alternative Medicine*, vol. 2020, no. 1, p. 7576818, 2020.

https://doi.org/10.1155/2020/7576818.

[20] T. Oo *et al.*, "Inhibition of Bacterial Efflux Pumps by Crude Extracts and Essential Oil from Myristica fragrans Houtt. (Nutmeg) Seeds against Methicillin-Resistant Staphylococcus aureus," *Molecules*, vol. 26, no. 15,

https://doi.org/10.3390/molecules26154662

[21] H. J. D. Dorman and S. G. Deans, "Antimicrobial agents from plants: antibacterial activity of plant volatile oils," *Journal of Applied Microbiology*, vol. 88, no. 2, p. 308-316, 2000.

https://doi.org/10.1046/j.1365-2672.2000.00969.x.

[22] S. S. A, T. S. Biju, A. P. Francis, G. R, V. P. Veeraraghavan, and K. Sankaran, "Fabrication of Biogenic Titanium Nanoparticles and Investigating Their Biological Properties for Dental Applications," (in eng), *Cureus*, vol. 15, no. 8, p. e44209, 2023.

https://doi.org/10.7759/cureus.44209.

[23] R. S. Bennion, J. E. Thompson, E. J. Baron, and S. M. Finegold, "Gangrenous and perforated appendicitis with peritonitis: treatment and bacteriology," (in eng), *Clinical therapeutics*, vol. 12 Suppl C, p. 31-44, 1990.

PMID: 2202510.

[24] E. F. Al-Jumaily and M. H. Al-Amiry, "Extraction and Purification of Terpenes from Nutmeg (myristica fragrans)," *Al-Nahrain Journal of Science*, vol. 15, no. 3, p. 151-160, 2012.

https://doi.org/10.22401/JNUS.15.3.21.

[25] A. D. Gupta, V. K. Bansal, V. Babu, and N. Maithil, "Chemistry, antioxidant and antimicrobial potential of nutmeg (Myristica fragrans Houtt)," *Journal of Genetic Engineering and Biotechnology*, vol. 11, no. 1, p. 25-31, 2013.

https://doi.org/10.1016/j.jgeb.2012.12.001.

[26] D. Abdalkader and F. Al-Saedi, "Antibacterial effect of different concentrations of zinc sulfate on multidrug resistant pathogenic bacteria," *Systematic Reviews in Pharmacy*, vol. 11, no. 3, p. 282, 2020.

https://doi.org/10.5530/srp.2020.3.32.

[27] M. T. Jasim, A. F. A. Alzubaidi, and S. M. D. Al-Rubaye, "Isolation and identification of Staphylococcus aureus and Pseudomonas aeruginosa bacteria from burns and wounds patients in Diyala governorate,"

International journal of health sciences, vol. 6, no. S9, p. 273-282, 2022.

https://doi.org/10.53730/ijhs.v6nS9.12222.

[28] A. R. Qader and J. A. Muhamad, "Nosocomial infection in sulaimani burn hospital, iraq," (in eng), *Ann Burns Fire Disasters*, vol. 23, no. 4, p. 177-81, 2010.

PMID: 21991221.

[29] J. M. R. Alshara, "Isolation and Identification of Bacterial Wound Infection Isolates and their Antibiotic Susceptibility Pattern," (in en), *Al-Kufa University Journal for Biology*, vol. 10, no. 2, p. 9-19, 2018.

https://doi.org/10.36320/ajb/v10.i2.8126.

[30] P. S. Gangania, V. A. Singh, and S. S. Ghimire, "Bacterial isolation and their antibiotic susceptibility pattern from post-operative wound infected patients," *Indian Journal of Microbiology Research*, vol. 2, no. 4, p. 231-235, 2015.

https://doi.org/10.5958/2394-5478.2015.00020.5.

[31] Y. S. Baek, J. Jeon, J. W. Ahn, and H. J. Song, "Antimicrobial resistance of Staphylococcus aureus isolated from skin infections and its implications in various clinical conditions in Korea," *International Journal of Dermatology*, vol. 55, no. 4, p. e191-e197, 2016.

https://doi.org/10.1111/ijd.13046.

[32] S. Chen, L. Rao, and C. Lin, "The dissemination of fusidic acid resistance among staphylococcus epidermidis clinical isolates in Wenzhou, China," *Infection and Drug Resistance*, p. 2537-2544, 2022.

https://doi.org/10.2147/IDR.S365071.

[33] M. B. Sannathimmappa, V. Nambiar, and R. Aravindakshan, "Antibiotic Resistance Pattern of Acinetobacter baumannii Strains: A Retrospective Study from Oman," *Saudi Journal of Medicine & Medical Sciences*, vol. 9, no. 3, 2021.

https://doi.org/10.4103/sjmms.sjmms_855_20.

[34] A. A. Al-Sultan, "Prevalence of High-Risk Antibiotic Resistant Acinetobacter baumannii in the Holy Cities of Makkah and Al-Madinah," *The Open Microbiology Journal*, vol. 15, no. 1, p. 145-151, 2021.

https://doi.org/10.2174/1874285802115010145.

[35] M. Al-Tamimi *et al.*, "Multidrug-Resistant Acinetobacter baumannii in Jordan," *Microorganisms*, vol. 10, no. 5,

https://doi.org/10.3390/microorganisms10050849

[36] T. Awoke *et al.*, "High Prevalence of Multidrug-Resistant Klebsiella pneumoniae in a Tertiary Care Hospital in Ethiopia," *Antibiotics*, vol. 10, no. 8,

https://doi.org/10.3390/antibiotics10081007

[37] A. Rashid Mahmood and N. Mansour Hussein, "Study of Antibiotic Resistant Genes in Pseudomonas aeroginosa Isolated from Burns and Wounds," (in en), *Archives of Razi Institute*, vol. 77, no. 1, p. 403-411, 2022.

https://doi.org/10.22092/ari.2021.356681.1893.

[38] M. Muddassir *et al.*, "A Study of Isolation and Identification of Multidrug Resistant Pseudomonas aeruginosa from Wound Specimen," *Journal of Pharmaceutical Research International*, vol. 32, no. 32, p. 26-31, 2020.

https://doi.org/10.9734/jpri/2020/v32i3230930.

[39] C. O. Nyambane, "Prevalence and susceptibility profiles of bacterial pathogens associated with urinary tract infections in children presenting at Kisii level 5 hospital, Kisii County, Kenya," MSc, Department of Microbiology, Kenyatta University, I56/CE/23495/2011, 2015.

https://ir-library.ku.ac.ke/items/d4dd7f6a-129a-4016-bdca-84c77c75ff72.

[40] Q. Qiu, G. Zhang, X. Sun, and X. Liu, "[Study on chemical constituents of the essential oil from Myristica fragrans Houtt. by supercritical fluid extraction and steam distillation]," (in chi), *Journal of Chinese Medicinal Materials*, vol. 27, no. 11, p. 823-826, 2004.

PMID: 15810588.

[41] M. T. Sultan *et al.*, "Nutritional and therapeutic potential of nutmeg (Myristica fragrans): A concurrent review," *Cogent Food & Agriculture*, vol. 9, no. 2, p. 2279701, 2023.

https://doi.org/10.1080/23311932.2023.2279701.

[42] X.-W. Yang, X. Huang, and M. Ahmat, "[New neolignan from seed of Myristica fragrans]," (in chi), *Journal of Chinese Materia Medica*, vol. 33, no. 4, p. 397-402, 2008.

PMID: 18533495

[43] Y. M. J. Taihuttu and R. D. Agustin, "Antioxidant and antibacterial activity of nutmeg oil (Myristica fragrans Houtt) from Ambon Moluccas," *AIP Conference Proceedings*, vol. 2588, no. 1, p. 020012, 2023.

https://doi.org/10.1063/5.0113251.

[44] B. Narasimhan and A. S. Dhake, "Antibacterial Principles from Myristica fragrans Seeds," *Journal of Medicinal Food*, vol. 9, no. 3, p. 395-399, 2006.

https://doi.org/10.1089/jmf.2006.9.395.

[45] T. Thileepan, V. Thevanesam, and S. Kathirgamanathar, "Antimicrobial activity of seeds and leaves of Myristica fragrans against multi-resistant microorganisms," *J. Agric. Sci. Technol. A*, vol. 7, p. 302-308, 2017.

https://doi.org/10.17265/2161-6256/2017.05.002.