EVALUATION OF GINGER AND GARLIC FOR ANTIMICROBIAL ACTIVITY AGAINST STAPHYLOCOCCUS AND STREPTOCOCCUS SPECIES ISOLATED FROM WOUND SAMPLES

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Abstract

This study aimed to evaluate the antimicrobial activity of ginger and garlic extracts against *Staphylococcus* and *Streptococcus* species isolated from wound samples collected from the surgical and orthopedic departments of ESUT Teaching Hospital. This is a cross-sectional study conducted between 15th of July to 20th of November 2024. A total of 100 patients were studied and data was collected using standard methods. Wound sample were cultured on Mannitol salt agar and Blood agar respectively. The ginger and garlic extract were processed and 20g of each was immersed in 100ml of 95% ethanol for 72hrs. Serial dilutions of different concentrations (100mg/ml, 200mg/ml and 400mg/ml) were prepared using phosphate buffered saline as a solvent. Susceptibility testing was done using agar well disc diffusion methods. Data was analyzed statistically using software SPSS version 25. *Staphylococcus aureus* 45(51.7%), *Staphylococcus epidermidis* 15(17.2%), *Streptococcus pyogenes* 20(23.0%), and *Streptococcus agalactiae* 7(8.1%). *Staphylococcus aureus* was the most isolated species, 25 (55.6%) of isolates in males and 20 (44.4%) in females. *Staphylococcus aureus* was most prevalent in the 21–30 age group with 12 isolates (26.7%), followed by the 11–20 and 31–40 age groups with 10 isolates each (22.2%), and fewer isolates in the 0–10 (8, 17.8%) and 41+ (5, 11.1%) groups. Ginger extract, the zones of inhibition ranged from 10.0 ± 1.2 mm to 19.2 ± 1.8 mm across the bacterial species, with the highest activity observed against *Staphylococcus aureus* at 400 mg/mL. Garlic extract the zones of inhibition ranged from 12.3 ± 1.3 mm to 21.3 ± 2.1 mm, also showing the greatest effect on *Staphylococcus aureus* at 400 mg/mL. Garlic exhibited higher zone than ginger with the most effect on 400mg/mL. The antimicrobial activity of the extracts was compared with conventional antibiotics (amoxicillin and ciprofloxacin), their potency was lower than that of the antibiotics tested. This study suggests that ginger and garlic extracts have potential antimicrobial properties that could complement conventional treatment options for wound infections, particularly those caused by *Staphylococcus* and *Streptococcus* species.

INTRODUCTION

Wound infections are a critical concern in healthcare settings due to their potential to cause significant morbidity and mortality. Among the pathogens responsible for these infections, *Staphylococcus aureus* (*S. aureus*) and *Streptococcus* species are among the most prevalent and problematic. *S. aureus* is a Gram-positive bacterium known for its ability to colonize the skin and mucous membranes of humans. It is particularly notorious for causing a range of infections, from minor skin infections to life-threatening conditions such as sepsis and endocarditis (Tong *et al.,* 2015). Similarly, *Streptococcus* species, particularly *Streptococcus pyogenes* (Group A *Streptococcus*), are also common culprits in wound infections, causing diseases that range from superficial skin infections to invasive conditions like necrotizing fasciitis and toxic shock syndrome (Bisno *et al.,* 2003).

The management of *S. aureus* and *Streptococcus* infections is complicated by their ability to develop resistance to multiple antibiotics. Methicillin-resistant *Staphylococcus aureus* (MRSA) and antibiotic-resistant Streptococcus strains represent significant public health challenges due to their resistance to beta-lactam antibiotics and other antimicrobial agents (Turner *et al.,* 2019; Efstratiou & Lamagni, 2016). The emergence of antibiotic-resistant strains of these bacteria has led to increased healthcare costs, longer hospital stays, and higher mortality rates (Wang *et al.,* 2018).

Given the challenges posed by antibiotic resistance, there is growing interest in exploring alternative antimicrobial agents. Natural products have been used for centuries in traditional medicine and are now being investigated for their potential as sources of new antimicrobial compounds. Among these natural products, ginger (Zingiber officinale) and garlic (Allium sativum) have garnered significant attention due to their historical use in treating various ailments and their reported antimicrobial properties (El-Saber Batiha *et al*., 2020).

Ginger contains bioactive compounds, including gingerol, which has shown antimicrobial activity against a variety of bacteria, including *S. aureus* and *Streptococcus* species (Singh *et al*., 2021). Similarly, garlic has been used for thousands of years for its medicinal properties. The primary bioactive compound in garlic, allicin, has been extensively studied for its broad-spectrum antimicrobial effects against bacterial strains such as *S. aureus* and *Streptococcus* species (Borlinghaus *et al.,* 2014).

This study aims to evaluate the antimicrobial susceptibility of *S. aureus* and *Streptococcus* species isolated from wound samples to extracts of ginger and garlic. The findings could provide insights into the potential use of these natural products as alternative or adjunctive treatments for bacterial wound infections, especially given the rising problem of antibiotic resistance.

Materials and Methods

Study Population: The study population included 100 patients recruited from the surgical and orthopedic wards of Enugu State University Teaching Hospital, Parklane.

Sample Collection: Wound samples were collected using sterile swabs from patients attending the hospital, and transported in a sterile medium for further analysis.

Isolation and Identification of Staphylococcus aureus and Streptococcus Species: Upon receipt, wound samples were cultured on Mannitol salt agar and blood agar for the isolation of *Staphylococcus* and *Streptococcus* species respectively.

Preparation of Ginger and Garlic Extracts: Fresh ginger and garlic were purchased from local markets, crushed, and soaked in 100ml of 95% ethanol for 72 hours for extraction. Twenty grams (20g) of garlic and ginger powder was soaked in 100ml of 95% ethanol for 72 hours at room temperature with intermittent agitation. The mixture was then filtered using Whatman No. 1 filter paper, and the filtrate was concentrated using a rotary evaporator at 40°C to obtain the crude extracts. The extracts were stored at 4°C to preserve the antibacterials property until use and standardized to a different concentration via serial dilution concentration by dissolving a specific amount (100mg/ml. 200mg/ml, 400mg/ml) of the extract in a measured volume of phosphate buffered saline for Ginger and Garlic.

Antimicrobial Susceptibility Testing: Antimicrobial susceptibility was determined using the agar well diffusion method, with standardized suspensions of each isolate being inoculated onto Mueller-Hinton agar. Wells of 6 mm diameter will be punched into the agar, and 100 ml, 200 ml and 400 ml of the ginger and garlic extracts were dispensed into each well. The plates were incubated at 37°C for 24 hours. After incubation, the zones of inhibition around the wells were measured to assess the antimicrobial activity.

Data Analysis: Data were analyzed statistically using SPSS version 25.

RESULTS

Table 4.1: Distribution of Bacterial Isolates

Bacterial Species	Number of Isolates	Percentage (%)
Staphylococcus aureus	45	51.7
Staphylococcus epidermidis	15	17.2
Streptococcus pyogenes	20	23.0
Streptococcus agalactiae	7	8.1
Total Positive	**87**	**100.0**
Negative Samples	13	-

Table 4.1 The distribution of bacterial isolates shows that *Staphylococcus aureus* was the most frequently isolated species, accounting for 51.7% of the total positive samples, followed by *Streptococcus pyogenes* at 23.0%. *Staphylococcus epidermidis* made up 17.2% of the isolates, while *Streptococcus agalactiae* was the least prevalent at 8.1%. Out of 100 total samples, 87 were positive for bacterial isolates, representing 87.0% of the samples, while the remaining 13.0% were negative. This indicates a high prevalence of bacterial pathogens, with *Staphylococcus aureus* being the dominant species among the isolates.

Table 4.2: Distribution of Bacterial Species by Gender

Bacterial Species	Male (n, %)	Female (n, %)	Total (n, %)	X^2	P value
Staphylococcus aureus	25 (55.6%)	20 (44.4%)	45 (51.7%)	1.362	0.431
Staphylococcus epidermidis	10 (66.7%)	5 (33.3%)	15 (17.2%)		
Streptococcus pyogenes	12 (60.0%)	8 (40.0%)	20 (23.0%)		
Streptococcus agalactiae	5 (71.4%)	2 (28.6%)	7 (8.1%)		
Total Positive	52 (59.8%)	35 (40.2%)	87 (100.0%)		

$P<0.05$ is significant, $p>0.05$ is not significant.

Table 4.2 shows the distribution of bacterial species among male and female participants. *Staphylococcus aureus* was the most commonly isolated species, accounting for 25 (55.6%) of isolates in males and 20 (44.4%) in females, with a total of 45 (51.7%). *Staphylococcus epidermidis* showed a higher proportion in males (10 isolates, 66.7%) compared to females (5 isolates, 33.3%), contributing to a total of 15 (17.2%). Similarly, *Streptococcus pyogenes* was more prevalent in males (12 isolates, 60.0%) than females (8 isolates, 40.0%), totaling 20 (23.0%). *Streptococcus agalactiae* had 5 isolates (71.4%) in males and 2 (28.6%) in females, making up 7 (8.1%). Overall, male participants accounted for a higher proportion of isolates (52, 59.8%) compared to females (35, 40.2%). There was no significant difference when compared between gender $p>0.05$

Table 4.3: Distribution of Bacterial Species by Age Group

Bacterial Species	0–10 (n, %)	11–20 (n, %)	21–30 (n, %)	31–40 (n, %)	41+ (n, %)	Total (n, %)	X^2	p-value
Staphylococcus aureus	8 (17.8%)	10 (22.2%)	12 (26.7%)	10 (22.2%)	5 (11.1%)	45 (51.7%)	0.974	0.071
Staphylococcus epidermidis	2 (13.3%)	3 (20.0%)	4 (26.7%)	4 (26.7%)	2 (13.3%)	15 (17.2%)		
Streptococcus pyogenes	3 (15.0%)	5 (25.0%)	6 (30.0%)	4 (20.0%)	2 (10.0%)	20 (23.0%)		
Streptococcus agalactiae	1 (14.3%)	2 (28.6%)	3 (42.9%)	1 (14.3%)	0 (0.0%)	7 (8.1%)		
Total Positive	14 (16.1%)	20 (23.0%)	25 (28.7%)	19 (21.8%)	9 (10.3%)	87 (100.0%)		

P<0.05 is significant, p>0.05 is not significant.

Table 4.3 presents the distribution of bacterial species across different age groups. *Staphylococcus aureus* was most prevalent in the 21–30 age group with 12 isolates (26.7%), followed by the 11–20 and 31–40 age groups with 10 isolates each (22.2%), and fewer isolates in the 0–10 (8, 17.8%) and 41+ (5, 11.1%) groups. *Staphylococcus epidermidis* showed the highest frequency in the 21–30 and 31–40 age groups with 4 isolates each (26.7%), and lower frequencies in the 0–10 (2, 13.3%), 11–20 (3, 20.0%), and 41+ (2, 13.3%) groups. *Streptococcus pyogenes* was most common in the 21–30 group with 6 isolates (30.0%) and progressively fewer in the 11–20 (5, 25.0%), 31–40 (4, 20.0%), 0–10 (3, 15.0%), and 41+ (2, 10.0%) groups. *Streptococcus agalactiae* was predominantly found in the 21–30 age group with 3 isolates (42.9%), followed by the 11–20 (2, 28.6%) and 0–10 and 31–40 (1 each, 14.3%) groups, with no isolates in the 41+ group. Overall, the 21–30 age group recorded the highest number of isolates (25, 28.7%), highlighting it as the most affected demographic.There was no significant difference when compared between different age groups p>0.05.

Table 4.4: General Characteristics of *Staphylococcus* and *Streptococcus* Isolates

Test/Observation	Staphylococcus	Streptococcus
Growth Medium	Mannitol Salt Agar	Blood Agar
Colonial Morphology	Yellow colonies (mannitol fermenting - *S. aureus*) or pink colonies (non-mannitol fermenting - *S. epidermidis*)	Small, pinpoint, translucent colonies with beta-hemolysis (*S. pyogenes*) or alpha-hemolysis (*S. agalactiae*)
Gram Stain	Gram-positive cocci in clusters	Gram-positive cocci in chains
Catalase Test	Positive	Negative
Coagulase Test	Positive for *S. aureus*, negative for *S. epidermidis*	Not Applicable

This table summarizes the observable characteristics and biochemical results of the bacterial isolates, providing critical information for their identification.

Table 4.5a: Antimicrobial Susceptibility of Ginger Extract

Bacterial Species	100 mg/mL	200 mg/mL	400 mg/mL
Staphylococcus aureus	12.5 ± 1.2	16.0 ± 1.6	19.2 ± 1.8
Staphylococcus epidermidis	11.8 ± 1.4	15.2 ± 1.8	18.0 ± 2.2
Streptococcus pyogenes	10.6 ± 1.3	14.3 ± 1.5	16.8 ± 1.7
Streptococcus agalactiae	10.0 ± 1.2	13.8 ± 1.3	15.6 ± 1.5

Table 4.5b: Antimicrobial Susceptibility of Garlic Extract

Bacterial Species	100 mg/mL	200 mg/mL	400 mg/mL
Staphylococcus aureus	14.8 ± 1.4	18.7 ± 1.9	21.3 ± 2.1
Staphylococcus epidermidis	13.6 ± 1.2	17.2 ± 1.6	19.7 ± 1.9
Streptococcus pyogenes	12.9 ± 1.1	16.8 ± 1.4	19.5 ± 1.6
Streptococcus agalactiae	12.3 ± 1.3	16.2 ± 1.5	18.9 ± 1.7

The antimicrobial susceptibility of ginger and garlic extracts, as shown in **Tables 4.5a and 4.5b,** indicates that the effectiveness of both extracts increases with concentration. For ginger extract, the zones of inhibition ranged from 10.0 ± 1.2 mm to 19.2 ± 1.8 mm across bacterial species, with the highest activity observed against *Staphylococcus aureus* at 400 mg/mL. Similarly, garlic extract demonstrated stronger antimicrobial activity, with inhibition zones ranging from 12.3 ± 1.3 mm to 21.3 ± 2.1 mm, also showing the greatest effect on *Staphylococcus aureus* at 400 mg/mL. Across all concentrations, garlic extract consistently exhibited greater antimicrobial activity than ginger extract against all tested bacterial species.

Table 4.6: Comparison of Extracts (400 mg/mL) with Conventional Antibiotics

Bacterial Species	Ginger Extract	Garlic Extract	Amoxicillin	Ciprofloxacin
Staphylococcus aureus	19.2 ± 1.8	21.3 ± 2.1	22.0 ± 1.5	25.3 ± 2.0
Staphylococcus epidermidis	18.0 ± 2.2	19.7 ± 1.9	20.5 ± 1.7	24.8 ± 1.8
Streptococcus pyogenes	16.8 ± 1.7	19.5 ± 1.6	21.3 ± 1.6	26.1 ± 2.1
Streptococcus agalactiae	15.6 ± 1.5	18.9 ± 1.7	20.0 ± 1.4	24.6 ± 1.7

Table 4.6 compares the antimicrobial efficacy of ginger and garlic extracts at 400 mg/mL with conventional antibiotics. While both extracts showed significant activity, their efficacy was slightly lower than that of amoxicillin and ciprofloxacin. Garlic extract at 400 mg/mL showed a closer performance to amoxicillin, particularly against *Staphylococcus aureus* (21.3 ± 2.1 mm) and *Streptococcus pyogenes* (19.5 ± 1.6 mm). Ciprofloxacin exhibited the highest zones of inhibition overall, highlighting its superior antibacterial efficacy. These findings emphasize that while natural extracts like ginger and garlic are effective antimicrobials, they may be less potent than conventional antibiotics at comparable concentrations.

DISCUSSION, CONCLUSION, AND RECOMMENDATIONS

DISCUSSION

This study evaluated the antimicrobial activity of ginger and garlic extracts against *Staphylococcus* and *Streptococcus* species isolated from wound samples. The findings revealed that *Staphylococcus aureus* was the most prevalent isolate, accounting for 51.7% of the bacterial pathogens identified. This dominance of *S. aureus* aligns with previous studies, such as Adegoke *et al*. (2020), which emphasized its frequent association with wound infections due to its ability to form biofilms and produce virulence factors. The second most common isolate, *Streptococcus* *pyogenes* (23.0%), was consistent with findings from Okeke *et al.* (2021), who reported its involvement in wound infections, particularly in post-surgical and trauma cases. The high prevalence of these pathogens underscores their clinical relevance and the need for effective antimicrobial agents.

Gender-based distribution revealed that males had a higher proportion of bacterial isolates compared to females across all species. *Staphylococcus aureus*, the most frequently isolated pathogen, accounted for 55.6% of isolates in males and 44.4% in females. Similarly, *Streptococcus pyogenes* was more common in males (60.0%) than females (40.0%), while *Staphylococcus epidermidis* and *Streptococcus agalactiae* also showed higher frequencies in male participants. This trend suggests that males may experience a higher risk of wound infections, possibly due to differences in occupation, lifestyle, or exposure to infection-prone environments.

Age-wise, the isolates were most prevalent in individuals aged 21–30, who accounted for 28.7% of all isolates. *Staphylococcus aureus* showed the highest frequency in this age group, followed by the 11–20 and 31–40 age brackets. Other species, including *Streptococcus pyogenes* and *Streptococcus agalactiae*, followed similar patterns, with decreasing frequencies in younger and older age groups. These findings highlight the increased susceptibility to bacterial infections among young adults, possibly due to higher physical activity levels, occupational exposure, or less stringent wound care practices.

The antimicrobial susceptibility results indicated that both ginger and garlic extracts exhibited inhibitory effects against all tested bacteria, with their effectiveness increasing with concentration. Garlic extract demonstrated higher zones of inhibition compared to ginger extract at all concentrations, with the greatest activity observed against *S. aureus* at 400 mg/mL (21.3 ± 2.1 mm). These findings are supported by Ojo *et al*. (2018) and Bello *et al.,* (2023), whose studies attributed garlic's superior efficacy to allicin, a compound with potent antimicrobial properties. Ginger extract, although less effective than garlic, showed promising activity, particularly at higher concentrations, aligning with Ibrahim *et al.* (2020), who highlighted the role of gingerols and shogaols in its antibacterial activity. Our Findings disagrees with Nabi *et al.,* (2022) where ginger showed the highest zone of inhibition against all bacterial isolates.

When compared with conventional antibiotics such as amoxicillin and ciprofloxacin, both ginger and garlic extracts exhibited significant but relatively lower efficacy. Ciprofloxacin showed the highest zones of inhibition overall, consistent with its broad-spectrum antibacterial activity and synthetic origin. However, garlic extract at 400 mg/mL demonstrated comparable activity to amoxicillin against *S. aureus* and *S. pyogenes*, suggesting its potential as a complementary antimicrobial agent. Similar observations were reported by Kumar *et al*. (2017), who noted garlic's capacity to act as a natural antibiotic alternative. Nabi *et al*., (2022) in their study found ginger to be more susceptible than conventional antimicrobials.

The difference in antimicrobial efficacy between the natural extracts and conventional antibiotics can be attributed to factors such as the purity of active components, extraction methods, and the bacterial resistance profile. Despite being less potent than synthetic antibiotics, garlic and ginger extracts present significant advantages, including their natural origin, reduced side effects, and lower likelihood of resistance development. This study reinforces the value of plant-based antimicrobials in managing wound infections caused by *Staphylococcus* and *Streptococcus* species, while also highlighting the need for tailored preventive and treatment strategies based on demographic patterns such as gender and age.

Conclusion

This study demonstrates that ginger and garlic extracts possess significant antimicrobial activity against *Staphylococcus* and *Streptococcus* species isolated from wound samples, with garlic extract exhibiting greater efficacy than ginger. *Staphylococcus aureus* was identified as the most prevalent species, particularly affecting male participants and individuals aged 21–30 years. While conventional antibiotics showed higher antibacterial activity, garlic extract at 400 mg/mL exhibited comparable efficacy to amoxicillin against *S. aureus* and *S. pyogenes*. These findings highlight the potential of natural plant extracts, particularly garlic, as complementary antimicrobial agents in managing wound infections.

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