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Antibacterial Activities of Some Medicinal Plants against *Escherichia coli, Vibrio* and *Salmonella* species isolated from seafood

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ABSTRACT

Seafood-associated bacterial contamination is an important food-safety concern, particularly for shellfish that are exposed to aquatic environments and may accumulate microorganisms. This study evaluated bacterial contamination of five selected shellfish—mussel, periwinkle, blood clam, rock snail and oyster—and assessed the antibacterial activity of ethanolic garlic (*Allium sativum*), neem (*Azadirachta indica*) and clove (*Syzygium aromaticum*) extracts against representative seafood-associated bacteria. Bacterial loads were determined from the outer surface, inner fluid and intestine, while agar-well diffusion and broth dilution were used to assess plant-extract activity and minimum inhibitory concentration (MIC), respectively. The highest total heterotrophic bacterial load on the outer surface was recorded in mussel ($7.3 \pm 0.2 \times 10^6$ CFU/cm²), while rock snail recorded the highest inner-fluid load ($9.0 \pm 0.2 \times 10^7$ CFU/mL). Among the target bacteria, *Vibrio* spp. was detected at the highest levels in the inner fluid of mussel (1.8×10^4 CFU/mL) and intestine of mussel (1.1×10^4 CFU/g). *E. coli* was detected on the outer surface of oyster (5.6×10^3 CFU/cm²) but was not detected in the inner fluid or intestine of the selected shellfish. Clove produced the greatest zones of inhibition across the concentration range, followed by garlic, whereas neem generally showed weaker activity. MIC values were 3.125, 3.125 and 6.25 mg/mL for clove against *E. coli*, *V. parahaemolyticus* and *S. enterica*, respectively; the corresponding values for garlic were 12.5, 6.25 and 25 mg/mL and for neem were 50, 25 and 50 mg/mL. The findings indicate substantial variation in bacterial contamination among shellfish and anatomical compartments and demonstrate promising in vitro antibacterial activity of clove and garlic extracts.

Keywords: Seafood, Garlic, Neem, Clove, Antibacterial Activity, Minimum Inhibitory Concentration, *E. coli*, *Vibrio*, *Salmonella.*

Introduction

Seafood is an important source of high-quality protein and micronutrients, but consumption is not without microbiological risk. Seafood-associated infections may result from contamination of harvesting waters, handling, processing and preparation, with the risk varying according to the commodity and its ecological characteristics (Iwamoto et al., 2010). Shellfish are of particular interest because filter-feeding bivalves can concentrate microorganisms present in surrounding waters. *Vibrio* spp. are naturally associated with coastal and estuarine environments and can contaminate seafood, especially shellfish.

Consumption of raw or inadequately cooked seafood is an established route of exposure to pathogenic *Vibrio* species (Baker-Austin et al., 2018). *Salmonella* and *Escherichia coli* are also important indicators or causes of foodborne contamination and may enter seafood through polluted waters or poor post-harvest handling (Iwamoto et al., 2010). Recent work in Port Harcourt also reported substantial bacterial contamination and multiple potential pathogens in selected seafood, emphasizing the continuing local food-safety concern (Amadi-Wali et al., 2025).

The search for natural antimicrobial agents has increased because plant materials contain diverse bioactive compounds.

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Garlic contains allicin and related sulfur-containing compounds with documented antibacterial activity (Choo et al., 2020). Clove is particularly rich in eugenol, a compound with broad antimicrobial activity against Gram-negative and Gram-positive bacteria and food-associated microorganisms (Marchese et al., 2017; Hu et al., 2018). Neem also has documented antimicrobial potential, although activity varies with plant part, extraction procedure and test organism (Wylie & Merrell, 2022).

Minimum inhibitory concentration provides a quantitative measure of antimicrobial activity by identifying the lowest concentration that prevents visible bacterial growth under defined test conditions (Wiegand et al., 2008; Kowalska-Krochmal & Dudek-Wicher, 2021). This study therefore focused on three anatomical parts of five species of shellfish and examined their bacterial loads alongside the antibacterial activity of garlic, neem and clove extracts.

Materials and Methods

Study Area

This study was carried out in Port Harcourt. Port Harcourt is the capital of Rivers State in the Niger Delta region of southern Nigeria. The city lies along the Bonny River and experiences a tropical monsoon climate characterized by high humidity throughout the year. Port Harcourt is an important center for fishing, seafood trading, marine transportation, and industrial activities. According to Nigerian Meteorological Agency (NIMET, 2020), the city experiences substantial rainfall and high humidity.

Collection of Medicinal Plants and Seafood Samples

Fresh neem leaves (*Azadirachta indica*), dried clove buds (*Syzygium aromaticum*) and fresh garlic bulbs (*Allium sativum*) were purchased from local sellers in Railway markets, Mile1 Diobu, Port Harcourt. They were carefully sorted to remove damaged portions and washed thoroughly under running tap water to remove dust. The samples were air-dried at room temperature, ground into fine powder using a clean electric grinder, placed in sterile labelled ziplock bags and stored until extraction (Okoliew & Ajuru, 2014).

Fresh shellfish species: mussel (*Galatea paradoxa*), periwinkle (*Tympanotonus fuscatus*), blood clam (*Tegillarca granosa*), rock snail (*Thaisella coronata*) and oyster (*Crassostrea tulipa*), were purchased from seafood markets in Port Harcourt, including Creek Road, Mile One and Ikpukulu waterfront, New Road, Borikiri. The samples were labelled, placed in polythene bags, transported in an ice-packed cooler, and delivered immediately to the laboratory for microbial analysis. The samples were processed within 24 hours of collection.

Collection of Samples from Shellfish and Bacterial examination

Samples were collected from three anatomical parts of the shellfish (Surface of the shell -outer surface; inner fluid in the mantle cavity and the content of the intestine). For the outer surface, a sterile cotton swab moistened with sterile normal saline (0.9% sodium chloride (NaCl) was swabbed over approximately 1cm^2 for smaller samples and 4 cm^2 for larger samples area of the sample surface. For the inner fluid, 1mL of internal fluid was aspirated from the mantle cavity using a sterile syringe and homogenized in sterile normal saline (1:10 w/v) for serial dilution and bacteriological examination. For the intestinal content, The intestine was carefully removed, opened longitudinally and approximately 1g of the content was collected with sterile loops and emulsified in sterile normal saline (1:10 w/v) for serial dilutions.

Total heterotrophic bacterial counts were determined using standard plate-count procedures and expressed as CFU/cm² for the outer surface, CFU/mL for inner fluid and CFU/g for intestine. Target bacteria were isolated using selective/differential media: TCBS agar for *Vibrio* spp., Salmonella-Shigella agar for *Salmonella* spp. and EMB agar for *E. coli*. Suspected colonies were characterized by standard biochemical tests (MacFaddin, 2000).

Preparation of plant extracts

Approximately 100 grams of each powdered plant material was transferred into sterile flasks and mixed with 300ml of 90% ethanol. The mixtures were allowed to stand for 72hrs, with intermittent agitation. Each mixture was filtered through Whatman No.1 filter paper (Whatman Ltd., England) using sterile funnel.

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The filtrates were concentrated using a rotary evaporator, and the remaining solvent was removed in a water bath maintained at 40^0C. The concentrated extracts were stored in sterile universal bottles until use (Abubakar & Haque, 2020). The Sterility of each plant extracts was assessed by inoculating 0.5 mL from each extract on sterile Mueller Hinton Agar plates and incubating at 37 ˚C for 24hrs. The absence of visible growth was interpreted as evidence that the extracts were sterile (CLSI, 2021). The extracts were diluted in two-fold with sterilized 5% Dimethyl Sulfoxide (DMSO) to obtain concentrations of 100, 50, 25, 12.5, 6.25, 3.125, 1.56, 0.78 mg/mL.

Antibacterial Assay

Agar well diffusion method according to Owusu *et al.,* (2021) was employed with slight modification to evaluate the antibacterial activity of crude ethanoic plant extract. Fresh cultures of respective pathogens (approximately 1×108 CFU/mL) were streaked evenly over the surface of the sterile Mueller Hinton agar plates with a sterile cotton swab; then, the plates were left to dry for ten minutes. After drying, wells were made on each plate using a sterilized cork borer measuring 6 mm in diameter. Then the eight concentrations of the test extracts were dispensed into the labelled wells. For the comparison, gentamicin (0.025 mg/mL) and 5% DMSO (100mg/mL well) were used as positive and negative controls, respectively.

The plates were then kept in the refrigerator for 2hrs to allow the extract to diffuse properly into the medium. The plates were then incubated at 37 °C for 24hrs, and the Zone of Inhibition (ZOI) was measured and recorded in millimeters. Antibacterial activities of crude extracts were interpreted by calculating the area of the inhibition zone around the well. Antimicrobial activity evaluation of each test plant was assessed by measuring the size of the inhibition zone to the nearest mm, and the values were expressed as the mean zone of inhibition at different concentrations ± standard error of the mean (SEM).

Molecular Identification

DNA extraction, DNA quantification, the 16srRNA Amplification, sequencing of the amplified product was carried out using the Big-Dye Terminator kit on a 3510 ABI sequencer by Inqaba Biotechnological, Pretoria South Africa. The bla_{CTX-M} genes from the bacterial isolates were amplified using the bla_{CTX-M}F: 5'-AAAAATCACTGCGCCAGTTC-3' and bla_{CTX-M}R: 5'-AGCTTATTCATCGCCACGTT-3' primers on ABI 9700 Applied Biosystems thermal cycler at a final volume of 40 micro-litres for 35 cycles. Plasmid DNA was extracted from the selected bacterial isolates using the Zyppy™ Plasmid Miniprep Kit (Catalog Nos. D4036) according to manufacturers' instructions.

Minimum inhibitory concentration

MIC was determined by broth dilution using two-fold serial concentrations. The MIC was defined as the lowest concentration at which visible bacterial growth was inhibited. The approach follows established dilution principles for MIC testing (Wiegand *et al.*, 2008; Kowalska-Krochmal & Dudek-Wicher, 2021).

Statistical analysis

Data were summarized using means, standard deviations and percentages. One-way analysis of variance to assess differences among extracts and concentrations at $p < 0.05$.

Results

The results of the Total heterotrophic bacterial (THB) count for the seafood samples is reported in Table 1, the outer surface showed mussel had the highest bacterial load ($7.3 \pm 0.2 \times 10^6$ CFU/cm^2), followed by oyster ($6.9 \pm 0.2 \times 10^6$ CFU/cm^2), blood clam ($1.9 \pm 0.01 \times 10^6$ CFU/cm^2) and rock snail ($1.7 \pm 0.01 \times 10^6$ CFU/cm^2) while periwinkle had the lowest contamination at $3.9 \pm 5.0 \times 10^5$ CFU/cm^2.

In the mantle (inner) fluid, highest bacterial load was seen with rock snail ($9.0 \pm 0.2 \times 10^7$ CFU/mL), followed by mussel, blood clam and oyster ($1.1 \pm 0.1 \times 10^7$ CFU/mL, $1.2 \pm 0.8 \times 10^7$ CFU/mL, and $1.2 \pm 0.1 \times 10^7$ CFU/mL) respectively. Least contamination was observed with Periwinkle ($8.5 \pm 0.5 \times 10^6$ CFU/mL).

In the intestine, blood clam and rock snails showed bacterial load of $1.2 \pm 0.01 \times 10^7$ CFU/g, mussels had bacterial load of $1.0 \pm 0.1 \times 10^7$ CFU/g, lowest contamination was seen with periwinkle ($7.3 \pm 0.3 \times 10^6$ CFU/g) and oyster ($6.8 \pm 0.5 \times 10^6$ CFU/g).

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Table 1: Total heterotrophic bacterial loads of the five selected shellfish in different anatomical compartments

Sample from Anatomical Part	Shellfish Samples				
	Mussel	Periwinkle	Blood clam	Rock snail	Oyster
OS (CFU/cm²)	$7.3 \pm 0.2 \times 10^6$	$3.9 \pm 5.0 \times 10^5$	$1.9 \pm 0.01 \times 10^6$	$1.7 \pm 0.01 \times 10^6$	$6.9 \pm 0.2 \times 10^6$
IF (CFU/mL)	$1.1 \pm 0.1 \times 10^7$	$8.5 \pm 0.5 \times 10^6$	$1.2 \pm 0.8 \times 10^7$	$9.0 \pm 0.2 \times 10^7$	$1.2 \pm 0.1 \times 10^7$
IN (CFU/g)	$1.0 \pm 0.1 \times 10^7$	$7.3 \pm 0.3 \times 10^6$	$1.2 \pm 0.01 \times 10^7$	$1.2 \pm 0.01 \times 10^7$	$6.8 \pm 0.5 \times 10^6$

The distribution of confirmed target bacterial isolate is presented in Table 2. Three target bacterial groups, namely *Vibrio* spp., *Salmonella* spp. and *Escherichia coli*, were detected among the seafood samples examined. *Vibrio* spp. had the highest number of positive recoveries 14 (50.0%), followed by *Salmonella* spp. 10 (35.7%) and *E. coli* 4(14.3%), making a total of twenty-eight-(outer surface (4), inner fluid (16), intestine (8) target bacterial isolates. *Vibrio* was represented by four seafood samples, *Salmonella* by four seafood samples and *E. coli* by one seafood samples. The species-level distribution shows that mussel and blood clam contributed the greatest numbers of *Vibrio* isolates, while oyster was the only selected shellfish from which *E. coli* was confirmed.

Table 2: Distribution of confirmed target-bacterial isolates among shellfish

Shellfish	*Vibrio* n (%)*	*Salmonella* n (%)*	*E. coli* n (%)*
Mussel	2 (14.3)	1 (10.0)	0 (0.0)
Periwinkle	0 (0.0)	1 (10.0)	0 (0.0)
Blood clam	2 (14.3)	0 (0.0)	0 (0.0)
Rock snail	1 (7.1)	1 (10.0)	0 (0.0)
Oyster	1 (7.1)	1 (10.0)	1 (25.0)

**Percentages are the percentages of the total isolates of each organism*

Table 3 revealed the results of target-bacterial loads by anatomical parts. The outer surface showed no detectable *E. coli* contamination among species, except for oyster that recorded 5.6×10^3 CFU/cm². *Vibrio* contamination was observed with oyster (5.9×10^3 CFU/cm²) > blood clam (3.8×10^2 CFU/cm²) > rock snail (2.4×10^2 CFU/cm²), no detectable contamination in periwinkle and mussel. *Salmonella* contamination was absent in mussel, periwinkle, and rock snail.

However, notable counts were seen with blood clam (4.5×10^2 CFU/cm²) and oyster (2.8×10^3 CFU/cm²).

E. coli was not detected in the inner fluid of any of the seafood species. *Vibrio* spp. were isolated from blood clam (6.9×10^3 CFU/ml), and oyster (6.8×10^3 CFU/ml), but absent in periwinkles. Highest contamination was seen in mussel (1.8×10^4 CFU/ml) while rock snail had the lowest count (3.8×10^3 CFU/ml). *Salmonella* contamination was present in all species, oysters recorded the highest count (4.5×10^3 CFU/ml), periwinkles, blood clam and rock snail, had a bacterial count of 3.5×10^3 CFU/ml each. Mussels recorded lowest *Salmonella* contamination (1.5×10^3 CFU/ml).

Table 3: Target-bacterial loads recorded in shellfish, by anatomical part

Organism/ Anatomical Part	Target-bacterial loads in Shellfish (Seafood)				
	Mussel	Periwinkle	Blood clam	Rock snail	Oyster
E. coli – OS (CFU/cm²)	0	0	0	0	5.6×10^3
Vibrio – OS (CFU/cm²)	0	0	3.8×10^2	2.4×10^2	5.9×10^3
Salmonella- OS (CFU/cm²)	0	0	4.5×10^2	0	2.8×10^3
Vibrio – inner fluid (CFU/mL)	1.8×10^4	0	6.9×10^3	3.8×10^3	6.8×10^3
Salmonella – inner fluid (CFU/mL)	1.53×10^3	3.5×10^3	3.5×10^3	3.5×10^3	4.5×10^3
Vibrio – intestine (CFU/g)	1.1×10^4	0	3.4×10^3	6.3×10^3	8.8×10^3
Salmonella – intestine (CFU/g)	6.0×10^3	0	3.3×10^3	0	4.6×10^3

- Outer surface (OS), inner fluid and intestine

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The intestine recorded *E. coli* was not detected in the intestine of any of the seafood species. However, Mussels showed highest *Vibrio* count (1.1×10^4 CFU/g), followed by oyster (8.8×10^3 CFU/g), moderate *Vibrio* contamination was observed with rock snail (6.3×10^3 CFU/g), while lowest contamination was seen with blood clam (3.4×10^3 CFU/g). No *Vibrio* contamination was seen in periwinkles. *Salmonella* showed moderate intestinal contamination, peaking in mussel (6.0×10^3 CFU/g) and oyster (4.6×10^3 CFU/g). Blood clam recorded lowest *Salmonella* contamination (3.3×10^3 CFU/g). Periwinkle and rock snail showed no detectable *Salmonella* contamination.

The molecular identification of five representative bacterial isolates was carried out using 16SrRNA gene amplification and sequencing. Agarose gel electrophoresis presented in Plate 1 showed successful amplification of the 16SrRNA gene in all five isolates, with distinct DNA bands corresponding to approximately 1,500 base pairs (bp), indicating successful Polymerase Chain Reaction (PCR) amplification.

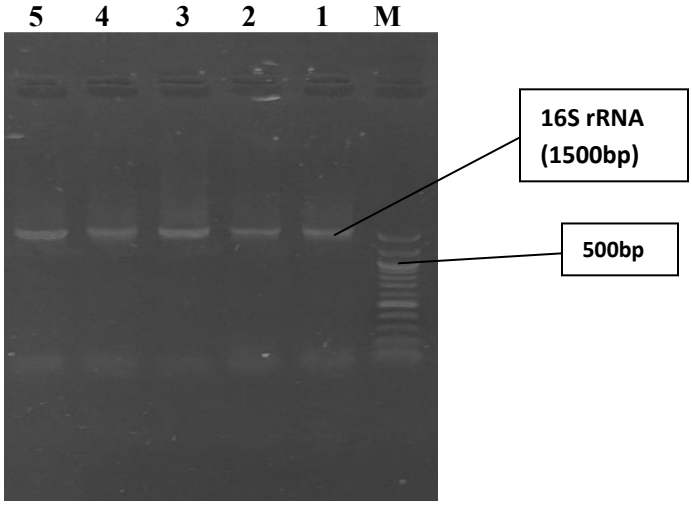

Plate 1: Amplified 16SrRNA gene fragments of approximately 1500 base pair (bp) from the selected bacterial isolates. Lane M represents the 100bp molecular marker, while lanes 1-5 represents the bacterial isolates.

The amplified gene sequences were subjected to BLAST analysis using the National Centre for Biotechnology Information (NCBI) non-redundant nucleotide database.

The results showed 100% sequence pairwise identity between the study isolates and referenced strains deposited in the data base. The isolates were identified as *Escherichia coli* strain U5/41 (accession numbers NR_024570.1), *Salmonella enterica subsp. enterica* strain AU08(accession numbers LR991664.1,), *Vibrio parahaemolyticus* strain NBRC 12711(accession numbers AB680329.1), *Vibrio vulnificus* ATCC 27562 (accession numbers U10862.1) and *Vibrio cholerae* strain KHS38 (accession numbers LC793860.1).

Plate 2 shows the amplification of the bla_{CTX-M} gene among the bacterial isolates the target gene, with an expected amplicon size of approximately 500 bp, was detected in *Escherichia coli* strain U5/41, *Vibrio parahaemolyticus* strain NBRC 12711, *Vibrio vulnificus* ATCC 27562 and *Vibrio cholerae* strain KHS38, whereas it was not detected in *Salmonella enterica subsp. enterica* strain AU08.

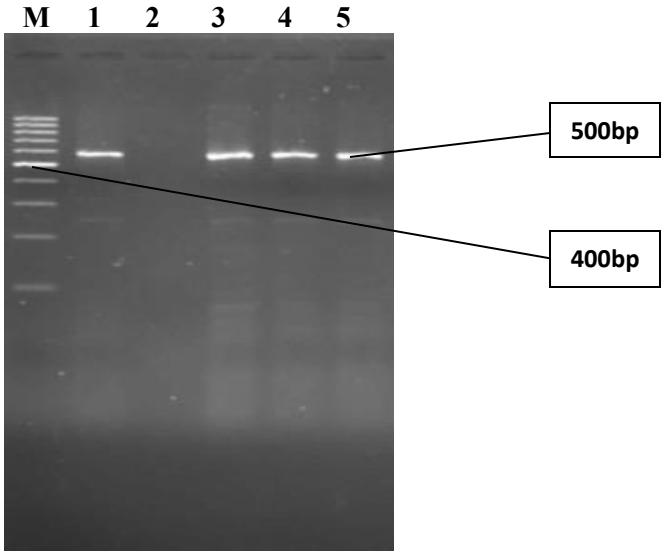

Plate 2: Amplified bla_{CTX-M} resistance gene bands. Lane 1-5 represents the bacterial isolates while M represents the 100bp molecular marker.

The antibacterial activities of the ethanolic extracts of garlic, neem and clove against the test organisms are presented in Table 4. The results show that clove extract produced the largest mean zone of inhibition against all test organisms at most of the concentrations evaluated. Garlic extract also demonstrated considerable antibacterial activity, producing moderate to high inhibition zones against *E. coli*, *Vibrio* spp., and *Salmonella* spp.

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Table 4: Mean inhibition zones produced by garlic, neem and clove extracts across tested concentration range

Concentration (mg/mL)	Plant Extract		
	Garlic (mm)	Neem (mm)	Clove (mm)
100	17.7	14.0	21.0
50	16.0	12.0	18.3
25	15.0	10.3	17.3
12.5	12.7	7.7	15.7
6.25	9.3	6.0	13.7
3.125	7.0	4.0	11.7
1.562	3.3	0.0	11.3
0.781	0.0	0.0	9.0

In contrast, neem extract exhibited the lowest antibacterial activity, with comparatively smaller inhibition zones across the concentrations tested.

The Minimum inhibitory concentration (MIC) result is presented in Table 5.

Table 5: Minimum inhibitory concentrations of garlic, neem and clove extracts against the representative bacterial isolates

Test organism	Garlic (mg/mL)	Neem (mg/mL)	Clove (mg/mL)
E. coli strain U5/41	12.5	50	3.125
V. parahaemolyticus strain NBRC 12711	6.25	25	3.125
S. enterica subsp. enterica strain AU08	25	50	6.25

The result revealed that clove extract recorded the lowest observed MIC value against *Escherichia coli* strain U5/41 (3.125 mg/ml), followed by garlic (12.5 mg/ml), while neem had the highest MIC (50 mg/ml). Against *Vibrio parahaemolyticus* strain *NBRC 12711*, clove extract produced the lowest observed MIC (3.125 mg/ml) values, followed by garlic (6.25 mg/ml), whereas neem recorded MIC value of 25 mg/ml. MIC value against *Salmonella enterica* subsp. enterica strain AU08 showed clove extract exhibited the lowest observed MIC (6.25 mg/ml), followed by garlic extract (25 mg/ml), while neem extract had the highest MIC (50 mg/ml).

Figure 1 clearly shows that clove had the lowest MIC values across all three organisms, followed by garlic, while neem had the highest MIC values.

Therefore, the graphical presentation supports the conclusion that clove extract showed greatest activity, followed by garlic and neem extracts in antibacterial effectiveness.

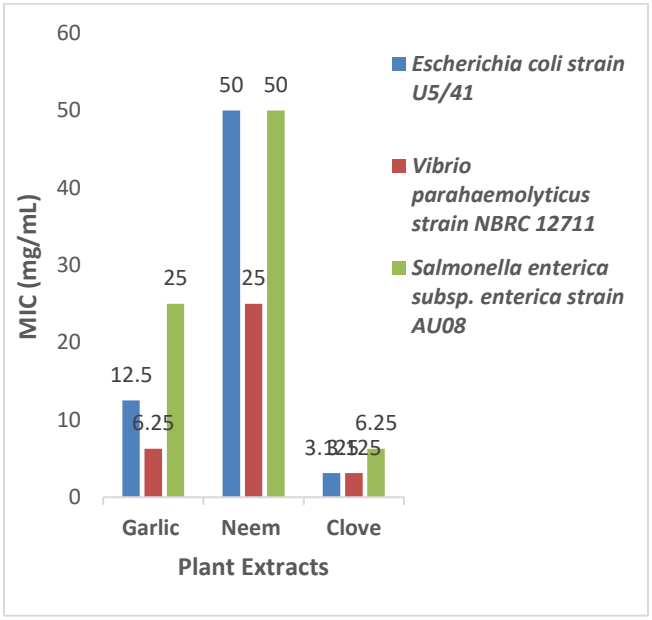

Figure 1: Minimum Inhibitory Concentration (MIC) of garlic, neem and clove against the three test organisms

Discussion

This study has revealed that, the total heterotrophic bacterial counts showed marked differences among the five shellfish and among anatomical parts. Mussel had the highest outer-surface count, whereas rock snail had the highest inner-fluid count. Such variation is biologically plausible because shellfish differ in feeding behaviour, habitat and exposure to sediments and surrounding water. Seafood-associated contamination is influenced by environmental conditions, harvesting practices and handling, and shellfish can present particular microbiological risks (Iwamoto et al., 2010). *Vibrio* was the most prominent target organism in the Group 2 results. The inner fluid of mussel had the highest *Vibrio* load (1.8×10^4 CFU/mL), followed by the intestine of oyster (8.8×10^3 CFU/g). The detection of *Vibrio* in several shellfish is consistent with the ecological distribution of *Vibrio* in marine and estuarine environments and the recognized association between shellfish consumption and vibriosis (Baker-Austin *et al.,* 2018).

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The result is also relevant to Port Harcourt, where Amadi-Wali *et al.* (2025) reported bacterial contamination and pathogenic organisms in selected seafood sold through local markets. *Salmonella* was detected in all three anatomical compartments across different shellfish, with the highest inner-fluid value occurring in oyster (4.5×10^3 CFU/mL) and the highest intestinal value occurring in oyster (4.6×10^3 CFU/g). *Salmonella* is not considered a normal component of marine seafood microbiota and its detection may indicate contamination from sewage, runoff or handling.

The finding is therefore of food-safety importance, particularly where shellfish are sold and prepared under conditions that may permit cross-contamination. *E. coli* was detected only on the outer surface of oyster among the five selected shellfish, at 5.6×10^3 CFU/cm², and was not detected in the inner fluid or intestine. Because *E. coli* is commonly used as an indicator of faecal contamination, its detection on oyster suggests possible exposure to contaminated water or handling surfaces. However, the result should not be generalized to all oysters because only the samples included in the present study were examined.

The plant-extract results showed a consistent potency order of clove > garlic > neem. Clove produced the largest inhibition zones from 100 to 0.781 mg/mL. This stronger activity is consistent with evidence that eugenol, a major constituent of clove oil, has broad antibacterial activity and can affect bacterial membranes and other cellular processes (Marchese *et al.,* 2017; Hu *et al.*, 2018). More recent reviews continue to support the potential of eugenol and clove-derived preparations against food-associated and resistant bacteria (Ribeiro *et al.,* 2024).

Garlic also showed measurable antibacterial activity and was more active than neem in the present assay. This finding is biologically plausible because allicin and related sulfur-containing compounds from garlic possess antimicrobial activity against a broad range of bacteria (Choo *et al.,* 2020).

Neem showed weaker activity in the present experiment, but this does not mean that neem lacks antibacterial potential. Reviews indicate that neem extracts and constituents can have antimicrobial effects, with activity influenced by the plant part, extraction method and target organism (Wylie & Merrell, 2022).

The MIC findings provide a quantitative comparison of the extracts. Clove had the lowest MIC against all three representative isolates: 3.125 mg/mL for *E. coli* and *V. parahaemolyticus* and 6.25 mg/mL for *S. enterica*.

Garlic required 12.5, 6.25 and 25 mg/mL, respectively, while neem required 50, 25 and 50 mg/mL. Lower MIC values indicate stronger inhibitory activity under the conditions of the assay (Wiegand *et al.*, 2008). The differences between extracts may reflect differences in the concentration and stability of their active constituents as well as bacterial susceptibility.

The present findings should be interpreted as in vitro evidence rather than proof that these extracts can safely preserve seafood. Plant-extract activity can change with extraction solvent, concentration, storage, food composition and contact conditions. Further work should therefore standardize the extracts, identify active constituents, test reproducibility and assess their effectiveness in real seafood matrices before application as food preservatives.

Conclusion

The five selected shellfish showed substantial variation in bacterial loads across the outer surface, inner fluid and intestine. *Vibrio* spp. occurred in several shellfish compartments and produced some of the highest target-organism counts recorded in the study, while *E. coli* was detected on the oyster outer surface. Among the three medicinal plant extracts, clove showed the strongest antibacterial activity, followed by garlic and neem. Clove also produced the lowest observed MIC values against the representative *E. coli*, *V. parahaemolyticus* and *S. enterica* isolates.

The findings provide preliminary information on the bacterial loads of the selected shellfish obtained from the sampled markets in Port Harcourt and support further investigation of clove and garlic as potential sources of natural antimicrobial compounds.

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