

Profiling of antimicrobial genes of pathogenic bacteria found in chicken sold in Port Harcourt, River State, Nigeria

Chika Christiana Nwankwo ^{1,*}, Chineye Nwogu ¹, Esther Oluomachi Umeh ¹ and Iheanyi Omezuruike Okonko ²

¹ Department of Microbiology Technology, School of Science Laboratory Technology, University of Port Harcourt, Choba, River State, Nigeria.

² Virus and Genomics Research Unit, Department of Microbiology, University of Port Harcourt, Choba, River State, Nigeria.

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Abstract

This study investigated the antimicrobial resistance (AMR) patterns and the distribution of antimicrobial-resistant genes (ARGs) and virulent genes in pathogenic bacteria isolated from raw, cooked and grilled chicken samples. Raw Chicken samples were bought from four cold rooms in Port Harcourt, Nigeria and examined for bacterial contamination. The grilled chicken samples were bought from street vendors. PCR amplification targeting virulent genes was used, and the bacterial isolates were identified molecularly. Eleven bacterial genera were isolated: *Streptococcus* sp, *Proteus vulgaris*, *Enterococcus faecalis*, *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Salmonella enterica*, *Escherichia coli*, *Enterobacter aerogenes*, *Acinetobacter*, *Listeria* spp., and *Staphylococcus aureus*. In the Raw and cooked samples, *Pseudomonas aeruginosa* (25%) had the highest percentage occurrence frequency, followed by *Salmonella enterica* (16.33%), *Escherichia coli* (16%), *Staphylococcus aureus* (12.24%), with *Proteus vulgaris* and *Acinetobacter* having the least. In the grilled sample, *Salmonella enterica* (22.4%) had the highest percentage occurrence, *Pseudomonas aeruginosa* (12.5%), *Streptococcus* sp (12.5%), *Escherichia coli* (10%), with *Listeria* sp and *Enterobacter aerogenes* having the least percentage of occurrence (6.12%). Multiple Antibiotic Resistance (MAR) ranged between 0.5 and 1.0 in the raw chicken sample. *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Enterococcus faecium* had high antibiotic resistance of >0.5, with *E. coli* having a MAR of 1.0, showing resistance to all 10 antibiotics studied. In the cooked chicken sample, the MAR ranged from 0.4 to 1.0, with *Acinetobacter* showing resistance to all 10 antibiotics studied. The grilled chicken has a MAR range of 0.2 to 1.0, with *Proteus vulgaris* having the highest multiple antibiotic resistance index of 1.0, with a resistance pattern to all antibiotics studied. Generally, it was observed that the pathogens showed high resistance to the commonly used antibiotics: Augmentin, Streptomycin, Ceporex, Ampiclox, and Ceftriaxone. *Salmonella* and *E. coli* persisted in cooked samples, which suggests potential risks for human health. PCR amplification of quinolone resistance-conferring gene (*qnrS*) genes in antibiotic-resistant bacteria isolated from grilled chicken meat revealed that *qnrS* genes were present in five out of the seven isolates, with an amplicon size of 395 bp. *Proteus vulgaris* and *Listeria* spp were negative for *qnrS* genes. Similarly, the *qnrA* gene was present in eight of nine pathogenic isolates screened. This study underscores the growing threat of AMR in foodborne pathogens, particularly in poultry. This makes treatment of infections caused by these bacteria more difficult and less effective, thus emphasising the need for improved food safety practices, stricter regulation of antibiotics in agriculture, and enhanced surveillance systems. Further research into alternative antimicrobial strategies is also recommended to mitigate the spread of resistant bacteria in the food chain.

Keywords: Antimicrobial Resistant genes; Antibiotic susceptibility; Pathogenic genes; PCR; Chicken

* Corresponding author: Chika C. NWANKWO, email: chika.nwankwo@uniport.edu.ng

1. Introduction

Antimicrobial resistance (AMR) is one of the top global public health and development threats. In 2019, it was estimated that bacterial AMR was directly responsible for 1.27million global deaths and contributed to 4.95million deaths [1]. The rise in antibiotic resistance poses a significant threat globally, reducing the efficacy of common antibiotics against widespread infections. In 2022, the Global Antimicrobial Resistance and Use Surveillance System (GLASS) report highlighted alarming rates among prevalent bacterial pathogens, with median rates in 76 countries of 42% for third-generation cephalosporin-resistant *E. coli* and 35% for Methicillin-resistant *Staphylococcus aureus*. In 2020, urinary tract infections caused by *E. coli*, 1 in 5 cases exhibited reduced susceptibility to standard antibiotics like ampicillin, co-trimoxazole and fluoroquinolones. Increased levels of resistance potentially lead to heightened utilisation of last-resort drugs like carbapenems, for which resistance is in turn being observed across multiple regions [50].

Antimicrobials have residues in chicken meat from Kiambu County, Kenya" Food Control, 2025 Publication, have been used in the agricultural sector as growth-promoting agents in sub-therapeutic dosages for feeding, improving feed conversion for high mass and product yield, and treating animal diseases and preventing their occurrence [28]. been used in subtherapeutic dosages in feeding as growth-promoting agents, improving feed conversion for high mass and product production, and treating and preventing illnesses in animals in the agricultural sector [28].

According to a recent study, the average yearly intake of antibiotics worldwide is 192 mg/PCU for pigs, 68 mg/PCU for hens, and 43 mg/PCU for cattle. This consumption is expected to rise from 93,000 tons in 2017 to 103,000 tons in 2030, which represents an 11.5% increase [48]. However, unfavourable effects, including the selection and spread of AMR during the course of antimicrobial use, pose serious concerns [27]. These concerns stem from the fact that, in developing nations, particularly those in Sub-Saharan Africa (SSA), a greater incorporation of sub-therapeutic doses in livestock husbandry, as a prophylactic measure, is becoming the norm, implying serious effects for both humans and animals [10].

Following a successful animal-to-human transmission, person-to-person transmission occurs within the human population. The uncontrolled use of antibiotics in farm animals is thought to exacerbate and maintain the emergence and spread of AMR in people and the environment [20].

Drug-resistant zoonotic infections may or may not have an impact on animal health, but may have a substantial impact on human health since the pathogens can be spread to people directly or indirectly through food, water, or the environment through leached animal waste and manure [26]. Many health issues in human communities, including high morbidity, chronic illnesses with high rates of recurrence, and high risks of disease metastasis, are caused by this transmission, which eventually lowers quality of life and, in many cases, results in death [5].

The overuse of antibiotics in the production of food animals is one of the contributing factors to antibiotic resistance, a global health concern. The expansion of farming operations in a large portion of the developing globe is predicted to result in a significant increase in usage in the upcoming years. Poultry is one of the most ubiquitous food sectors worldwide and chicken is the most extensively farmed species, with over 90 billion tons of chicken meat produced per year. In most nations, poultry are raised using a wide variety of antibiotics, primarily orally, to prevent or treat illness while simultaneously promoting growth and production.

Antibiotic-resistant genes in pathogens are likely to emerge more quickly as a result of the careless use of antibiotics in animal husbandry. The existence of antibiotic-resistant virulent gene residues in chicken raises concerns for human health in addition to the issues caused by the introduction of ARG in bacteria from poultry production. While poultry is a major source of protein globally, it also acts as a major vehicle for foodborne pathogens that can have an impact on public health. *Salmonella*, *Campylobacter*, *Escherichia coli*, and *Listeria monocytogenes* are among the common foodborne pathogens associated with poultry products, and they can cause serious foodborne illnesses [38]. During processing, handling, ingredient mixing, and exposure to airborne microorganisms can contaminate grilled chicken, which can lead to gastroenteritis and other serious health conditions when consumed [45].

2. Materials and methods

2.1. Sample Collection

A total of 16 raw chicken meat samples (thigh muscles, breast muscles, and liver) and 12 grilled chicken samples from different grocery stores and street vendors around Choba and Aluu, respectively, in Port Harcourt metropolis of River

State, Nigeria. The samples were collected in sterile zip-lock bags, labelled, and immediately placed in an ice box and transported to Microbiology Research Laboratory for analysis. All the samples collected were within the required date for consumption.

2.2. Sterilisation of Materials

All materials (media and glassware) used for this research work were autoclaved for 15 minutes at 121°C and 15psi for sterilization. The work benches were disinfected with 70% alcohol. Aseptic technique was maintained throughout the bench work. Flamed where needed.

2.3. Media Preparation

The preparation of media followed the protocol specified by the manufacturer which was used for isolation of bacteria in the chicken.

2.4. Analysis of samples

Standard autoclaving technique was used to sterilise the distilled water, culture media and glassware. Ten grams of each sample was added to 90 ml normal saline and homogenised for 2 minutes in a blender to make a solution. Each sample was serially diluted in 10-fold increments by adding 1.0 ml of homogenised sample to 9.0 ml sterile normal saline (0.85 % NaCl) and diluted from 10⁻¹ to 10⁻⁶. Standard microbiological techniques were employed. Macconkey agar, Centrimide agar, Nutrient agar and Mannitol salt agar was used to isolate possible pathogens and incubated for 24 hours at 37 °C. Characteristic colonies were counted and back-calculated as CFU/g in log scale [41]. The isolates were sub-cultured for another 24 hours, followed by a battery of biochemical tests, and observed traits were recorded. The colonial and morphological characterisation was done following Chesbrough's (2006) instructions. Bergeys manual of Systematic Bacteriology was also used to identify the isolates. Furthermore, Molecular analysis was carried out using the polymerase chain reaction to amplify DNA for the identification of the pathogens. The disk diffusion (Kirby- Bauer) method was carried out to determine the susceptibility of isolated pathogens to a panel of antibiotics commonly used in human and veterinary medicine.

2.5. Identification of Isolates

Salmonella, *Shigella*, *Proteus* and *E. coli* were isolated and identified using the methods and procedures recommended by standard methods [12] and [49]. For all organisms, selenite faeces (SF) broth was used as the primary enrichment medium at first. Each organism was then isolated using a different selective medium: *Salmonella-Shigella* (SS) agar for *Shigella* and *Salmonella*, Eosin Methylene Blue (EMB) agar for *E. coli*, and Blood agar for *Proteus*. In order to isolate *E. coli*, the inoculated media were first incubated for 24 hours at 37°C. During this time, special attention was paid to the unique colony characteristics, specifically the presence of a metallic green sheen on EMB agar, which is characteristic of *E. coli*. Colonies exhibiting these suspected features were then moved to nutrient agar and incubated for another 24 hours at 37°C for additional subculturing. A series of tests was performed for the initial identification, starting with the Gram stain and oxidase test. Following these initial assessments, additional biochemical analyses were performed, including tests for indole production, methyl red, Voges-Proskauer (VP), and the citrate utilisation test. In order to isolate and identify *Shigella* and *Salmonella*, enriched samples were cultivated on SS agar plates and cultured for 24 hours at 37°C. The development of a black core within the colony raised suspicions of *Salmonella*. Several biochemical assays, such as the Triple Sugar Iron agar (TSI) test, the indole test, the citrate test, and the urease test, were conducted to verify the identity of *Salmonella* species. Certain colonies were identified as *Salmonella* because they had an acid (yellow) butt and an alkaline (red) slant, produced hydrogen sulfide (H₂S) that caused the TSI agar to turn black, tested negative for tryptophan utilisation (shown by a yellow-brown ring in the indole test), and were negative for urea utilisation. Lastly, enriched samples were utilised to inoculate Blood agar plates in order to isolate and identify *Proteus mirabilis*. After that, these plated cultures were cultured for 24 hours at 37°C. Isolates were identified using normal microbiological procedures, which included morphological observations and evaluation of biochemical traits as outlined by [14].

2.6. Testing for Antimicrobial Susceptibility

Before conducting the susceptibility test, the inoculum was standardised using the guidelines provided by the Clinical and Laboratory Standards Institute (CLSI) in [19]. Using the Kirby-Bauer disk diffusion method on Mueller-Hinton Agar (Hi-Media Ltd., India), the antibiotic sensitivity pattern of bacterial isolates was determined in accordance with the Clinical and Laboratory Standards Institute (CLSI) guidelines 2020. Antibiotic discs were positioned on the agar plates using a disc dispenser and carefully pushed to guarantee full contact with the agar following sub-culturing on Mueller-Hinton agar. To test the sensitivity, the plates were then inverted and incubated for 24 hours at 37°C. Ten distinct

antibiotic discs were used: Ampicillin (10µg), Cephalexin (30µg), Ofloxacin (5µg), Gentamycin (10µg), Augmentin (30µg), Ciprofloxacin (5µg), Tetracycline (30µg), Streptomycin (30µg), and Pefloxacin (5µg). A ruler was used to measure the inhibitory zones' diameter after incubation, and the results were interpreted in accordance with CLSI recommendations [18].

2.7. Quality Control

Already typed-culture (*Escherichia coli* ATCC 25922 and *Staphylococcus aureus* ATCC 25923) was used as quality control for antimicrobial susceptibility testing as recommended by Clinical and Laboratory Standards Institute [14]

2.8. Detection of virulence genes

Following identification of the bacterial isolates, detection of the virulent genes of the isolates by PCR was done as described by other studies [44]. The PCR primers used for the amplification of the virulent genes are listed in Table 1. Briefly, bacterial cells from overnight cultures were suspended in 1ml distilled water and boiled at 95 °C for 10 minutes to obtain bacterial extracts. After boiling, the cell extracts were cooled on ice and immediately used as a DNA template for the PCR reaction. The PCR was done using the Phusion Flash High Fidelity PCR master mix (Finnzymes, Finland). The reactions were performed in a final volume of 10 µl containing 5µl phusion flash PCR master mix, 0.5µM of primer sets in 1µl volume of each and 2 µl of PCR water. The Piko thermal cycler (Finnzymes Instruments Oy, Finland) was programmed at 95 °C for 30 seconds for initial denaturation, followed by 35 cycles consisting of 95 °C for one second, 58 °C for 5 seconds and 72 °C for 15 seconds. Final extension was given at 72 °C for 1 minute. Detection was identified by the presence of a specific DNA band on 1.5 % agarose gel, stained with ethidium bromide and evaluated under a UV transilluminator. The estimation of the sizes of the PCR products was done according to the migration pattern of a 100-bp ladder.

2.9. PCR-based confirmation of Antibiotic-Resistant Genes (ARGs)

PCR amplification was carried out against ten ARGs with the bacterial DNA. The presence of genes associated with resistance to antibiotics was determined by PCR and the set of primers used for each gene is shown in Table 2. PCR reactions were performed in a total volume of 25 µl, including 1.5mM MgCl₂, 50mM KCl, 10mM Tris-HCl (pH 9.0), 0.1% Triton X-100, 200 µm of each dNTP (Fermentas), 1 µm primers, 1 IU of Taq DNA polymerase (Fermentas), and 5 µl (40–260 ng/µl) of DNA. Amplification reactions were carried out using a DNA thermo-cycler (Eppendorf Mastercycler, Eppendorf-Nethel-Hinz GmbH, Hamburg, Germany) as follows: Three min at 95 °C, 35 cycles each consisting of 1 min at 94 °C, 90 s at ~55 °C and 1 min at 72 °C, followed by a final extension step of 10 min at 72 °C. Amplified samples were analyzed by electrophoresis in 1.5% agarose gel and stained by ethidium bromide. A molecular weight marker with 100 bp increments (100 bp DNA ladder, Fermentas) was used as a size standard. Strains of *E. coli* O157:K88ac: H19, CAPM 5933 and *E. coli* O159:H20, CAPM 6006 were used as positive controls.

Table 1 PCR-based confirmation of Antibiotic-Resistant Genes (ARGs)

Target organism	Gene	Primers	Sequences 5'-3'	Size (bp) of PCR product	Reference
<i>Staphylococcus aureus</i>	<i>Sea</i>	Forward	TTGGAAACGGTTAAAACGAA	120	Armany <i>et al.</i> [7]
		Reverse	GAACCTTCCCATCAAAAACA		
<i>Escherichia coli</i>	<i>stx1</i>	Forward	CTT CGG TAT CCT ATT CCCGG	484	Mohammed & El Dahshan [33]
		Reverse	GGA TGCATC TCTGGT CAT TG		
<i>Pseudomonas aeruginosa</i>	<i>ToxA</i>	Forward	GACAACGCCCTCAGCATCACCAGC	396	Sharma & Das [44]
		Reverse	CGCTGGCCCATTCGCTCCAGCGCT		
<i>Salmonella</i> spp.	<i>invA</i>	Forward	GTGAAATTATCGCCACGTTCTGGGCAA	284	Mohammed & El Dahshan [33]
		Reverse	TCATCGCACCGTCAAAGGAACC		

Table 2: Primers used in this study to amplify antibiotic-resistant genes

Group of antibiotics	Target gene	Direction	Primer Sequence	Product length	Reference
Beta-lactams	bla _{KPC}	Forward	CATTCAAGGGCTTTCTTGCTGC	538	Racewicz et al. [42]
		Reverse	ACGACGGCATAGTCATTTGC		
	bla _{TEM}	Forward	CGCCGCATACACTATTCTCAG AATGA	445	
		Reverse	ACGCTCACCGGCTCCAGA TTTAT		
	bla _{NDM}	Forward	GGGCAGTCGCTTCCAACGGT	476	
Fluoroquinolones	qnrA	Forward	ATTTCTCACGCCAGGATTTG	529	Patel et al. [3]
		Reverse	GCAGATCGGCATAGCTGAAG		
	qnrB	Forward	GGMATHGAAATTCGCCACTG	429	
		Reverse	TTYGCBGYCGCCAGTCGAA		
	qnrS	Forward	GACGTGCTAACTTGCGTGAT	393	
		Reverse	TGGCATTGTTGGAAACTTG		
Aminoglycosides	strA-strB	Forward	ATGGTGGACCCTAAAACCTCT	893	
		Reverse	CGTCTAGGATCGAGACAAAG		
	aphA1	Forward	ATGGGCTCGCGATAATGTC	600	
		Reverse	CTCACCGAGGCAGTTCCAT		
	Aac(3)-II	Forward	ATATCGCGATGCATACGCGG	877	
		Reverse	GACGGCCTCTAACCGGAAGG		
Sulfonamides	sul1	Forward	CGGCGTGGGCTACCTGAACG	433	
		Reverse	GCCGATCGCGTGAAGTTCCG		
	sul2	Forward	CGGCATCGTCAACATAACCT	721	
		Reverse	TGTGCGGATGAAGTCAGCTC		
	sul3	Forward	CAACGGAAGTGGGCGTTG TGGA	244	
		Reverse	GCTGCACCAATTTCGCTGAACG		
Trimethoprim	dfr1	Forward	TGGTAGCTATATCGAAGAATG GAGT	425	
		Reverse	TATGTTAGAGGCGAAGTCTTG GGTA		
	dfr5	Forward	AGCTACTCTTTAAAGCCTTGA CGTA		
		Reverse	GTGTTGCTCAAAAACAACCTCG		
	dfr7/17	Forward	ACATTTGACTCTATGGGTGTT CTTC		
		Reverse	AAAACGTTCAAAAACCAAAT TGAA		
Tetracyclines	tetA	Forward	GGCGGTCTTCTTCATCATGC	502	Kocúřeková et al. [54]

		Reverse	CGGCAGGCAGAGCAAGTAGA		
	tetB	Forward	CGCCCAGTGCTGTTGTTGTC	173	
		Reverse	CGCGTTGAGAAGCTGAGGTG		
	tetC	Forward	GCTGTAGGCATAGGCTTGGT	888	
		Reverse	GCCGGAAGCGAGAAGAATCA		
Class 1 integrase	IntI1	Forward	GCCTTGCTGTTCTTCTACGG	565	Nirdesh et al. [55]
		Reverse	GATGCCTGCTTGTCTACGG		
Class 2 integrase	IntI2	Forward	AAGCAGACTTGACCTGA	565	
		Reverse	CACGGATATGCGACAAAAAGGT		

3. Results

3.1. Total Viable Counts of Bacteria from Raw and Cooked Chicken

Total viable counts of bacteria from raw and cooked chicken are presented in Table 3. the result revealed that the Total Heterotrophic Bacteria (THB) count for raw and cooked chicken was 0.47×10^5 cfu/g and 0.24×10^5 cfu/g. For the raw chicken, bacterial counts of 0.20×10^5 cfu/g, 23.5×10^5 cfu/g and 0.05×10^5 cfu/g were obtained from Centrimide agar, MacConkey agar and Mannitol Salt agar respectively. While for the the cooked chicken, bacterial counts of 0.20×10^5 cfu/g, 0.13×10^5 cfu/g, and 1.11×10^5 cfu/g were obtained from Centrimide agar, MacConkey agar and mannitol Salt agar respectively. Total viable count from grilled chicken revealed that the Total Heterotrophic Bacterial (THB) for grilled chicken is 34.0×10^5 cfu/g. A bacterial count of 16.85×10^5 cfu/g, 37.60×10^5 cfu/g, and 28.0×10^5 cfu/g were obtained from Centrimide agar, Macconkey agar and Mannitol Salt agar, respectively. Generally, the grilled chicken had the highest microbial load when compared to the cooked and raw chicken.

Table 3 Total viable counts of bacteria from raw and cooked chicken

Type of Bacterial count	Mean $\pm$ SE (cfu/g $\times 10^5$)		
	Raw chicken	Cooked chicken	Grilled Chicken
THB	0.47 ± 0.06	0.24 ± 0.00	34.0 ± 0.22
CA agar	0.20 ± 2.2	0.20 ± 3.61	16.85 ± 3.60
MAC agar	23.5 ± 0.05	0.13 ± 0.01	37.60 ± 0.51
MSA agar	0.05 ± 0.15	1.11 ± 0.10	28.0 ± 0.83

Counts represent means of triplicate samples $\pm$ standard error of samples

Key: cfu/g= Colony forming units per gramme; S.E=Standard error, NG= No growth; CA=centrimide agar; MAC=MacConkey agar; MSA=Mannitol salt agar

3.2. Percentage Occurrence of Bacterial Isolates from Raw and Cooked Chicken

The percentage occurrences of bacteria isolate from raw and cooked chicken are presented in Figure 1 reveals that *Pseudomonas aeruginosa* had the highest occurrence of 24.49% while *Bacillus cereus*, *Klebsiella pneumoniae*, *Acinetobacter* spp., *Proteus vulgaris* and *Corynebacterium* spp. were the least with equal occurrence of 6.12%. The percentage occurrence of bacterial isolates from grilled chicken (Table 4) showed that *Salmonella enterica* had the highest occurrence of (22.45%) while *Listeria* spp. and *Enterobacter aerogenes* were the least with equal occurrence of 6.12%. Furthermore, *Bacillus subtilis*, *Escherichia coli*, *Proteus vulgaris* and *Enterococcus faecalis* had equal occurrence of 10.20% while *Pseudomonas aeruginosa* and *Streptococcus* spp. had equal occurrence of 12.25%.

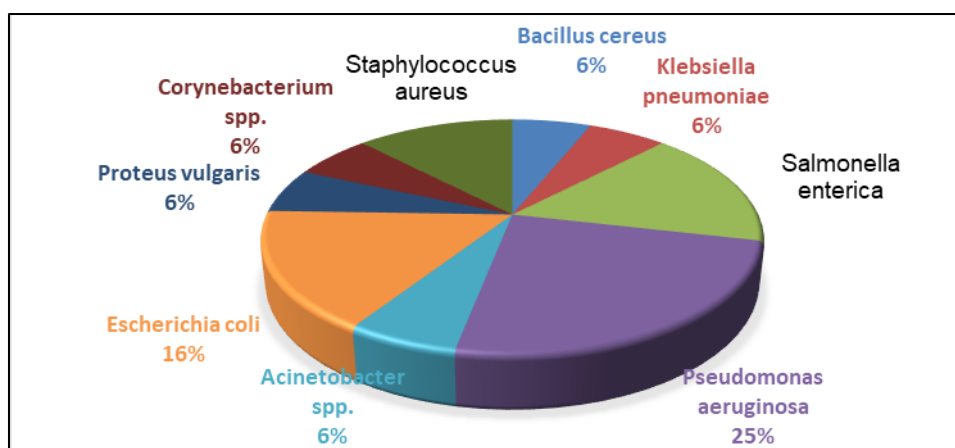


Figure 1 Percentage occurrence of bacterial isolates from raw and cooked meat chicken

Table 4 Percentage occurrence of bacterial isolates from grilled chicken

Bacterial isolate	Number of isolates	Occurrence (%)
<i>Bacillus subtilis</i>	5	10.20
<i>Salmonella enterica</i>	11	22.45
<i>Escherichia coli</i>	5	10.20
<i>Pseudomonas aeruginosa</i>	6	12.25
<i>Proteus vulgaris</i>	5	10.20
<i>Enterococcus faecalis</i>	5	10.20
<i>Streptococcus</i> spp.	6	12.25
<i>Listeria</i> spp.	3	6.12
<i>Enterobacter aerogenes</i>	3	6.12
Total	49	100

3.3. Antibiotic Resistance bacterial isolates from raw and cooked chicken

The growth of bacterial isolates in the presence of antibiotics is presented in Table 5 for Gram positive bacteria and Table 6 for Gram negative bacteria. Gram-positive bacterial isolates, *Bacillus cereus* and *Staphylococcus aureus* were found to be resistant to nine (9) out of ten (10) antibiotics while *Corynebacterium* spp. exhibited resistance to six (6) out of ten (10) antibiotics. These Gram-negative organisms *Escherichia coli* and *Acinetobacter* spp. were found to be resistant to ten (10) out of ten (10) antibiotics while *Salmonella enterica* and *Klebsiella pneumoniae* only showed resistant to four (4) out of ten (10) antibiotics.

Table 5 Antimicrobial susceptibility test for Gram-positive bacterial isolates from Raw chicken

Chicken type	Bacterial isolates	CPX	AZ	LEV	E	PEF	CN	APX	Z	AM	R
Raw	<i>Bacillus cereus</i>	S	22	20	19	18	17	R	R	R	15
Raw	<i>Enterococcus faecium</i>	18	20	S	20	20	R	16	R	S	22
Raw	<i>Corynebacterium</i> spp.	S	18	S	15	20	R	15	20	S	S
Raw	<i>Staphylococcus aureus</i>	S	22	20	22	20	R	R	15	R	22

PEF=Pefloxacin; CN=Gentamicin; APX=Ampiclox; Z=Zinnacef; AM=Amoxicillin; R= Rocephin; CPX=Ciprofloxacin; AZ=Azithromycin; LEV=Levofloxacin; E=Erythromycin; TRX=Ceftriaxone; CEP=Ceporex; CTZ=Ceftazidime. S=sensitive; R=Resistant, Zone of inhibition≤15mm (sensitive); Zone of inhibition≥15mm (Resistant)

Table 6 Antimicrobial susceptibility test for Gram-negative bacterial isolates from cooked and raw chicken

Code	Bacterial isolates	CEF	OFX	AU	PEF	CTZ	CN	CPX	CEP	TRX	S
Cooked	<i>Klebsiella pneumoniae</i>	S	S	20	S	S	S	12	R	R	R
cooked	<i>Salmonella enterica</i>	S	S	S	15	S	S	R	R	17	R
Cooked	<i>Salmonella enterica</i>	S	S	S	15	S	S	16	R	R	R
Raw	<i>Pseudomonas aeruginosa</i>	R	20	20	18	17	20	20	22	R	11
Raw	<i>Pseudomonas aeruginosa</i>	R	R	R	R	R	R	R	R	12	18
Cooked	<i>Acinetobacter</i> spp.	R	R	R	R	R	R	R	R	15	16
Raw	<i>Escherichia coli</i>	R	R	R	R	R	20	R	R	18	20
Raw	<i>Proteus vulgaris</i>	S	S	14	S	S	S	16	R	R	R

Keys: PEF=Pefloxacin; OFX= Travid; S=Streptomycin; SXT=Septrin; CH=chloramphenicol; SP=Sparfloxacin; CPX=Ciprofloxacin; AM=Amoxicillin; AU=Augmentin; CN=Gentamicin; TRX=Ceftriaxone; CEP=Ceporex; CTZ=Ceftazidime. S= Sensitive; R=Resistance; Zone of inhibition ≤ 15 mm (sensitive); Zone of inhibition ≥ 15 mm (Resistant).

The MAR index of raw chicken as shown in Table 7 ranged from 0.5 to 1.0 with *Escherichia coli* showing the highest resistance of 1.0 to all antibiotics (Ceporex, Perfloxin, Augmentin, Travid, Ceftazidime, Gentamicin, Ceftriaxone, sparfloxacin and Ciprofloxacin, this is followed by *Bacillus cereus*, *Staphylococcus aureus*, *Pseudomonas aeruginosa* (0.9) *Pseudomonas aeruginosa* was resistant to all except Ceftriazone, *Enterococcus faecium* (0.8 and) *Corynebacterium* spp (0.6), *Proteus vulgaris* (0.5). the pathogens showed various antibiotics resistance as shown in Table 7. The MAR of cooked chicken ranged from 0.4 to 1.0, with *Acinetobacter* spp having the highest MAR of 1.0 to all antibiotics used, while *Salmonella enterica* and *Klebsiella pneumonia* had a MAR of 0.4 respectively. For Gram-positive isolates from raw and cooked chicken as presented in Table 8, *Bacillus cereus* and *Staphylococcus aureus* exhibited resistance to nine of the ten antibiotics tested, posing significant treatment challenges for infections caused by these pathogens. Table 9 revealed high resistance rates among with Gram-negative bacteria such as *Escherichia coli* and *Acinetobacter* spp which showed resistance to all tested antibiotics, including ciprofloxacin, augmentin, and gentamicin. The MAR index values for these isolates (1.0) underscore their multidrug-resistant nature. In contrast, *Salmonella enterica* and *Klebsiella pneumoniae* exhibited resistance to fewer antibiotics, with MAR indices of 0.4, suggesting variability in resistance mechanisms among bacterial species.

Table 7 Multiple antibiotics resistance index of the bacterial isolates from Raw and cooked chicken

Code	Bacterial Isolates	Resistance pattern	No. of antibiotics	MAR Index
Cooked	<i>Salmonella enterica</i>	CPX, CEP, SP, TRX	4	0.4
Cooked	<i>Salmonella enterica</i>	CPX, CEP, SP, TRX	4	0.4
Raw	<i>Pseudomonas aeruginosa</i>	CEF, OFX, AU, PEF, CTZ, CN, CPX, CEP, TRX	9	0.9
Raw	<i>Pseudomonas aeruginosa</i>	CEF, OFX, AU, PEF, CTZ, CN, CPX, CEP, SP	9	0.9
Cooked	<i>Acinetobacter</i> spp.	CEF, OFX, AU, PEF, CTZ, CN, CPX, CEP, TRX, SP	10	1.0
Raw	<i>Escherichia coli</i>	CEF, OFX, AU, PEF, CTZ, CN, CPX, CEP, TRX, SP	10	1.0
Raw	<i>Bacillus cereus</i>	AZ, LEV, E, PEF, CN, APX, Z, AM, R	9	0.9
Cooked	<i>Klebsiella pneumonia</i>	AU, CEP, TRX, SP	4	0.4
Raw	<i>Proteus vulgaris</i>	CPX, CEP, TRX, SP	5	0.5
Raw	<i>Enterococcus faecium</i>	CPX, AZ, E, PEF, CN, APX, Z, R	8	0.8

Raw	<i>Corynebacterium</i> spp.	AZ, E, PEF, CN, CPX, Z	6	0.6
Raw	<i>Staphylococcus aureus</i>	AZ, LEV, E, PEF, CN, CPX, Z, AM, R	9	0.9

PEF=pefloxacin; CN=gentamicin; APX=ampiclox; Z=zinnacef; AM=Amoxicillin; R= Rocephin; CPX=Ciprofloxacin; AZ=Azithromycin; LEV=levofloxacin; E=Erythromycin; AU= augmentin; PEF=pefloxacin; OFX= Travid; S=Streptomycin; SP=Sparfloxacin; SXT=Septtrin; CH=chloramphenicol; CN= Gentamicin TRX=Ceftriaxone; CEP=Ceporex; CTZ=Ceftazidime.

3.4. Antibiotic susceptibility test for the Gram-positive bacterial isolates and Gram- negative from grilled chicken

Antimicrobial susceptibility test for Gram positive and Gram-negative bacterial isolates is indicated in Tables 10 and 11. The result for Gram positive revealed high rate of resistance to Amplicox and Azithromycin and high rate of susceptibility to Levofloxacin, Ceporex and Pefloxacin while for Gram negative it indicated a high rate of resistance to Ceftriaxone and Streptomycin and high-rate susceptibility to Pefloxacin, Ceporex and Gentamicin.

Table 8 Antimicrobial susceptibility test for Gram-positive bacterial isolates from Raw meat

Chicken type	Bacterial isolates	CPX	AZ	LEV	E	PEF	CN	APX	Z	AM	R
Raw	<i>Bacillus cereus</i>	S	22	20	19	18	17	R	R	R	15
Raw	<i>Enterococcus faecium</i>	18	20	S	20	20	R	16	R	S	22
Raw	<i>Corynebacterium</i> spp.	S	18	S	15	20	R	15	20	S	S
Raw	<i>Staphylococcus aureus</i>	S	22	20	22	20	R	R	15	R	22

PEF=pefloxacin; CN=gentamicin; APX=ampiclox; Z=zinnacef; AM=Amoxicillin; R= Rocephin; CPX=Ciprofloxacin; AZ=Azithromycin; LEV=levofloxacin; E=Erythromycin; TRX=Ceftriaxone; CEP=Ceporex; CTZ=Ceftazidime. S=sensitive; R=Resistant, Zone of inhibition≤15mm (sensitive); Zone of inhibition≥15mm (Resistant)

Table 9 Antimicrobial susceptibility test for Gram-negative bacterial isolates

Chicken type	Bacterial isolates	CEF	OFX	AU	PEF	CTZ	CN	CPX	CEP	TRX	S
Cooked	<i>Klebsiella pneumoniae</i>	S	S	20	S	S	S	12	R	R	R
Cooked	<i>Salmonella enterica</i>	S	S	S	15	S	S	R	R	17	R
Cooked	<i>Salmonella enterica</i>	S	S	S	15	S	S	16	R	R	R
Raw	<i>Pseudomonas aeruginosa</i>	R	20	20	18	17	20	20	22	R	11
Raw	<i>Pseudomonas aeruginosa</i>	R	R	R	R	R	R	R	R	12	18
Cooked	<i>Acinetobacter</i> spp.	R	R	R	R	R	R	R	R	15	16
Raw	<i>Escherichia coli</i>	R	R	R	R	R	20	R	R	18	20
Raw	<i>Proteus vulgaris</i>	S	S	14	S	S	S	16	R	R	R

PEF=pefloxacin; OFX= Travid; S=Streptomycin; SXT=Septtrin; CH=chloramphenicol; SP=Sparfloxacin; CPX=Ciprofloxacin; AM=Amoxicillin; AU= Augmentin; CN= Gentamicin; S= Sensitive; R=Resistance; Zone of inhibition≤15mm (sensitive); Zone of inhibition≥15mm (Resistant)

Table 10 Antibiotic susceptibility test for the Gram-positive bacterial isolates from grilled chicken

Code	Bacteria isolate	CPX	AZ	LEV	E	PEF	CN	APX	Z	AM	R
EG1	<i>Bacillus subtilis</i>	S	R	S	13	S	R	R	13	17	S
EG6	<i>Enterococcus faecalis</i>	S	R	S	10	S	R	R	11	20	S
EG7	<i>Streptococcus</i> spp.	S	S	S	17	S	18	R	17	R	18
EG8	<i>Listeria</i> spp.	22	R	S	R	S	R	R	12	R	R

CPX=Ciprofloxacin; AZ=Azithromycin; LEV=Levofloxacin; E=Erythromycin; PEF=Pefloxacin; CN=Gentamicin; APX=Ampiclox; Z=Zinnacef; AM=Amoxicillin; R= Rocephin; TRX=Ceftriaxone; CEP=Ceporex; CTZ=Ceftazidime. S=sensitive; R=Resistant, Zone of inhibition ≤15mm (sensitive); Zone of inhibition ≥15mm (Resistant).

Table 11 Antibiotic Susceptibility test for the Gram-negative and Gram-positive Bacterial isolates from Grilled Chicken

Code	Bacteria isolate	CEF	OFX	AU	PEF	CTZ	CN	CPX	CEP	TRX	S
EG1	<i>Bacillus subtilis</i>	R	21	20	S	R	S	S	S	R	R
EG2	<i>Salmonella enterica</i>	12	S	S	S	S	S	S	S	R	R
EG3	<i>Escherichia coli</i>	15	S	17	S	S	S	S	17	R	R
EG4	<i>Pseudomonas aeruginosa</i>	R	S	S	S	17	S	S	20	R	R
EG5	<i>Proteus vulgaris</i>	R	R	R	R	R	18	15	R	R	R
EG10	<i>Enterobacter aerogenes</i>	15	21	15	20	R	16	S	16	R	15
EG4	<i>Salmonella enterica</i>	R	23	15	17	14	20	S	15	R	15

CPX=Ciprofloxacin; AZ=Azithromycin; LEV=Levofloxacin; E=Erythromycin; PEF=Pefloxacin; CN=Gentamicin; APX=Ampiclox; Z=Zinnacef; AM=Amoxicillin; R= Rocephin; TRX=Ceftriaxone; CEP=Ceporex; CTZ=Ceftazidime. S=sensitive; R=Resistant, Zone of inhibition ≤ 15 mm (sensitive); Zone of inhibition ≥ 15 mm (Resistant).

3.5. Multiple Antibiotic resistance index of bacterial isolates from grilled chicken

Multiple antibiotic resistance index of bacterial isolates from grilled chicken are shown in Table 12. The result revealed that *Proteus vulgaris* had the highest multiple antibiotic resistance index of 1.0 with a resistance pattern to Ciprofloxacin, Ofloxacin, Augmentin, Pefloxacin, Ceftazidime, Gentamicin, Ciprofloxacin, Ceporex, Ceftriaxone and Streptomycin, while *Bacillus subtilis* and *Salmonella enterica* had the lowest multiple antibiotic indices with equal occurrence of (0.2). *Bacillus subtilis* has a resistance pattern to Ceporex, Ofloxacin, Augmentin, Ceftazidime, Ceftriaxone while *Salmonella enterica* has a resistance pattern to Ceftriaxone and Streptomycin. *Enterobacter aerogenes* were resistant to all antibiotics except ciprofloxacin, *Salmonella enterica*, *Listeria* spp and *Streptococcus* spp.

Table 12 Multiple antibiotics resistance index of the bacterial isolates of grilled chicken

Code	Bacterial Isolates	Resistance pattern	No. of antibiotics	MAR Index
EG1	<i>Bacillus subtilis</i>	CEF, OFX, AU, CTZ, TRX, S	2	0.2
EG2	<i>Salmonella enterica</i>	TRX, S	2	0.2
EG3	<i>Escherichia coli</i>	CEF, CEP, TRX, S	4	0.4
EG4	<i>Pseudomonas aeruginosa</i>	CTZ, CEP, TRX, S	4	0.4
EG5	<i>Proteus vulgaris</i>	CEF, OFX, AU, PEF, CTZ, CN, CPX, CEP, TRX, S	10	1.0
EG9	<i>Enterobacter aerogenes</i>	CEF, OFX, AU, PEF, CTZ, CN, CEP, TRX, S	9	0.9
EG10	<i>Salmonella enterica</i>	CEF, OFX, AU, PEF, CN, CEP, TRX, S	8	0.8
EG6	<i>Enterococcus faecalis</i>	AZ, CN, APX, AM	4	0.4
EG7	<i>Streptococcus</i> spp.	E, CN, APX, Z, AMR	6	0.6
EG8	<i>Listeria</i> spp.	CPX, AZ, E, CN, APX, AM, R	7	0.7

PEF=Pefloxacin; CN=Gentamicin; APX=Ampiclox; Z=Zinnacef; AM=Amoxicillin; R= Rocephin; CPX=Ciprofloxacin; AZ=Azithromycin; LEV=Levofloxacin; E=Erythromycin; AU=Augmentin; OFX=Ofloxacin; S=Streptomycin; TRX=Ceftriaxone; CEP=Ceporex; CTZ=Ceftazidime.

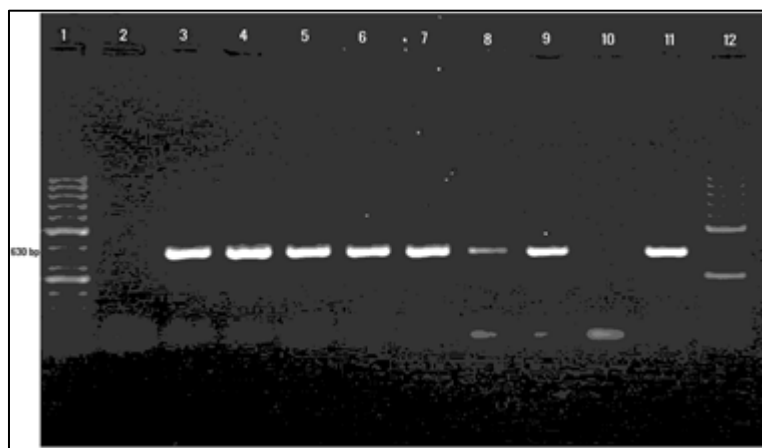
3.6. Molecular Characterisation of Resistance-Confering Gene in Antibiotic-Resistant Bacteria Isolated from Raw and Cooked Chicken Meat

Figure 2 presents the PCR amplification of the quinolone resistance-conferring gene (*qnrA*) in antibiotic-resistant bacteria isolated from raw and cooked chicken meat. Nine antibiotic-resistant bacteria were screened for *qnrA* genes. The gel electrophoresis revealed that *qnrA* genes were present in 8 out of the 9 isolates with an amplicon size of 630 bp. However, *Corynebacterium* sp, represented by lane 9, was negative for *qnrA* genes.

Figure 3 presents the PCR amplification of the quinolone resistance-conferring gene (*qnrS*) in antibiotic-resistant bacteria isolated from grilled chicken meat. Seven antibiotic-resistant bacteria were subjected to the *qnrS* genes. The

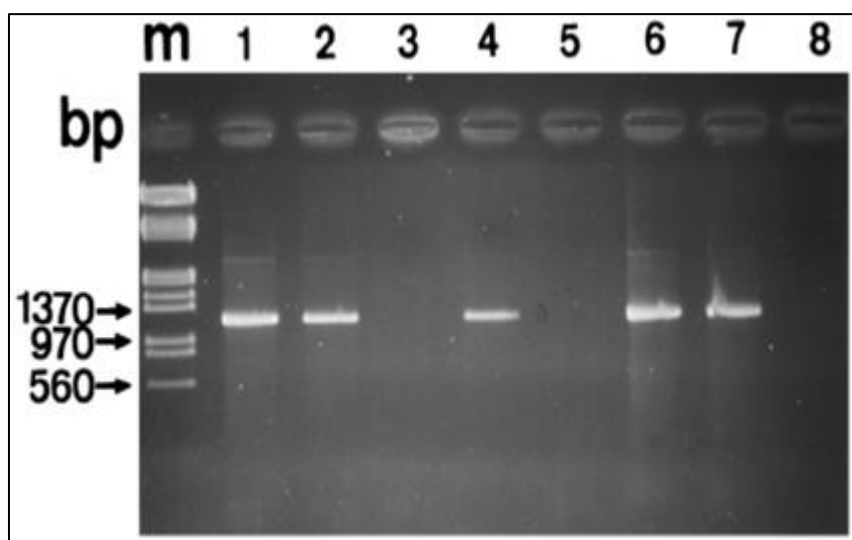
gel electrophoresis revealed that *qnrS* genes were present in 5 out of the 7 isolates with an amplicon size of 1395 bp. However, *Proteus vulgaris* and *Listeria* spp. represented by lanes 3 and 5 were negative for *qnrS* genes.

Most of the isolates studied harboured the *qnrS* and *qnrA* genes. As a result, the *qnrS* gene is suggested for PCR as an indicator for a rapid and reliable detection method from the chicken sample. Virulence genes are genes that allow bacteria to cause disease. They can encode factors that help bacteria attach to cells, invade tissues, produce toxins, and evade the immune system. Bacteria carrying virulence genes are more likely to cause severe infections.



Keys: Lane M=100 bp DNA marker, Lanes 1= Negative control, 2= *Bacillus cereus* (CN1), 3= *Klebsiella pneumoniae* (CN2), 4= *Pseudomonas aeruginosa* (CN5), 5= *Acinetobacter* sp. (CN7), 6= *Escherichia coli* (CN8), 7= *Proteus vulgaris* (CN9), 8= *Enterococcus faecium* (CN10), 9= *Corynebacterium* sp. (CN11), 10= *Staphylococcus aureus* (CN12)

Figure 2 Gel Electrophoresis Amplification of *qnrA* Genes in Antibiotic-Resistant Bacteria Isolated from Chicken Meat



Keys: Lane M=100 bp DNA marker, 1= *Bacillus subtilis*, 2= *Escherichia coli*, 3= *Proteus vulgaris*, 4= *Enterococcus faecalis*, *Streptococcus* spp., 5= *Listeria* spp., 6= *Enterobacter aerogenes*, 7= *Salmonella enterica*, 8= Negative control

Figure 3 Gel Electrophoresis Amplification of *qnrS* Genes in Antibiotic-Resistant Bacteria Isolated from Grilled Chicken

4. Discussion

The result revealed that the highest Total Heterotrophic Bacteria (THB) counts for raw and cooked chicken were 0.47×10^5 cfu/g and 0.24×10^5 cfu/g, respectively. respectively, this shows that the microbial load in the raw chicken is higher than that in the cooked chicken; this is similar to the findings of [36], who studied the microbial load of raw and cooked periwinkle and found that heat reduced the microbial load. However, the Total viable count from street-vended grilled chicken revealed a higher total heterotrophic bacteria load of 34.0×10^5 cfu/g when compared to the raw

and cooked chicken. This agrees with the work of [37][21] and [6], who reported that the processing method could be responsible for high microbial load, where a higher microbial load was recorded in street-vended grilled pork and beef. In the cooked chicken, the highest microbial load of 1.11×10^5 cfu/g was found in the Mannitol agar when compared to the counts obtained from Centrimide (0.20×10^5 cfu/g) and Manitol agar (0.05×10^5 cfu/g), respectively. The findings suggest that the handling process of cooked meat is a possible source of contamination even after the meat is cooked. The high level of counts in mannitol may be a result of a lack of personal hygiene of the processors, from dust and lack of good handling and manufacturing practices [29].

In the grilled chicken, bacterial loads of (16.85×10^5 cfu/g), (37.60×10^5 cfu/g), and (28.0×10^5 cfu/g) were obtained from Centrimide agar, MacConkey agar and Mannitol Salt agar, respectively. Generally, the grilled chicken had the highest microbial load when compared to the cooked and raw chicken. This agrees with previous works [6, 21], which reported that the processing method could be responsible for high microbial load. The identification of both Gram-positive and Gram-negative bacteria highlights the complex microbial ecosystem in chicken, particularly in raw samples, where conditions favour bacterial proliferation and diversity. A reduction in total heterotrophic bacteria in cooked chicken demonstrates the impact of cooking on bacterial load. However, the presence of viable bacteria, including pathogens such as *Salmonella* spp. and *Escherichia coli*, even in cooked samples, indicates faecal contamination during slaughter operations, evisceration, potential limitations of standard cooking practices, as well as poor personal hygiene. This finding reinforces the need for proper handling, thorough cooking, and avoidance of cross-contamination during food preparation. According to the Food and Agriculture Organisation of the United Nations (FAO), the risk of food poisoning associated with street food remains a threat in many parts of the world, with microbiological contamination being one of the major problems.

The raw, cooked and grilled chicken samples analysed in this study were contaminated with pathogenic bacteria. The following pathogenic bacteria were predominant in the raw and cooked samples: *Bacillus* spp., *Klebsiella* spp., *Salmonella* spp., *Pseudomonas aeruginosa*, *Acinetobacter* spp., *Escherichia* spp., *Proteus vulgaris*, *Enterococcus* spp., *Corynebacterium* spp. and *Staphylococcus* spp, with *Pseudomonas aeruginosa* (24.49%) as the highest percentage of frequency of occurrence in the raw and cooked samples, followed by *Salmonella enterica* (16.33%) and *Escherichia coli* (16%). In poultry environments, *Pseudomonas* species are commonly found in processing facilities and on raw poultry due to their high tolerance to various environmental conditions [22]. These bacteria can spoil poultry products, producing biofilms that aid in survival and resistance to sanitation efforts [30].

Furthermore, in the grilled chicken, *Salmonella enterica*, *Pseudomonas aeruginosa*, *Streptococcus* spp., *Bacillus subtilis*, *Escherichia coli*, *Proteus vulgaris*, and *Enterococcus faecalis* were predominantly found, with *Salmonella enterica* (22.5%) having the highest frequency of occurrence. The prevalence of *Salmonella enterica* in the present study (22.45%) was similar to that of a previous study [24], which isolated different species of *Salmonella* from raw and frozen chicken. This is also similar to reports from other parts of the world (25%, chicken meat [34] and the province of Iran (20%, chicken meat [32]. *Staphylococcus aureus*, *Escherichia coli*, *Salmonella enterica*, *Pseudomonas aeruginosa*, and *Streptococcus* species were also isolated from grilled meat and pork [20], and these pathogens were similar to those isolated from the chicken samples studied. Pathogens such as *Salmonella*, *Campylobacter*, *Escherichia coli*, and *Listeria monocytogenes* are commonly associated with poultry products, and they can cause severe foodborne illnesses [38]. These pathogens are often transmitted to humans through the consumption of undercooked or contaminated poultry, leading to gastroenteritis and other serious health conditions [45].

Studies have shown that *Salmonella* can persist in poultry farms and processing plants, and contamination often occurs during slaughter and handling [8]. In many countries, *Salmonella* infections from poultry products continue to be a leading cause of foodborne illness, despite strict biosecurity measures and regulatory interventions [31]. Contamination of poultry products with *Listeria* is often attributed to inadequate sanitation in processing environments. It can survive in biofilms on equipment and surfaces, allowing it to persist in food production facilities and pose a threat to food safety [13]. Meat containing the *E. coli* indicator organism is likely to be contaminated with faeces and indicates poor hygiene [47].

As an indicator of hygiene and sanitary quality, the presence of *E. coli* suggests that consumers are at risk of being food poisoned and the presence of other pathogenic fungi. Meat containing the *E. coli* indicator organism is likely to be contaminated with faeces and indicates poor hygiene [18]. Notably is the fact that *Enterobacter* species are bacteria commonly known to cause gastroenteritis, meningitis, and infection in the bladder [47]. More so, an enterotoxigenic strain of *E. coli* is the most common cause of traveller's diarrhoea, and some strains of this pathogen can cause a wide variety of infections, such as other forms of diarrhoea and other gastrointestinal problems, especially in a community setting [11]. Pork or other food products that contain *E. coli* in its infective dose can be a continuous source of infections leading to complications and death, especially among children and immunocompromised individuals [2].

Misuse and overuse of antimicrobials lead to antimicrobial resistance (AMR), a major global health concern [11, 18]. Antimicrobial Resistance (AMR) has become a big threat to global health. It has risen to dangerously high levels in all parts of the world, making it difficult to treat infectious diseases [3, 25, 40, 43]. Bacteria isolated from grilled chicken often exhibit multidrug resistance, meaning they are resistant to multiple classes of antibiotics. This makes treatment of infections caused by these bacteria more difficult and less effective. These pathogenic bacteria can increase the risk of food-borne illness, which can lead to hospitalisation and even death.

Escherichia coli, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Enterococcus faecium* have high antibiotic resistance of >0.5 in raw chicken, with *E. coli* having a MAR of 1.0. *E. coli* showed resistance to all 10 antibiotics studied in the raw samples. This agrees with the previous finding [9]. Poultry is a known reservoir for antibiotic-resistant *E. coli*, including strains that carry extended-spectrum beta-lactamases (ESBLs). These enzymes make *E. coli* resistant to cephalosporins, and ESBL-producing *E. coli* is frequently isolated from poultry products, heightening public health concerns [9]. *Pseudomonas* spp. in poultry is often multidrug-resistant, with resistance to beta-lactams, aminoglycosides, and fluoroquinolones. This resistance is associated with efflux pumps and beta-lactamase enzymes [23].

In the cooked chicken sample, *Acinetobacter* showed resistance to all 10 antibiotics studied: Ceporex, Ceftriaxone, Sparfloxacin, Ciprofloxacin, gentamicin, Ceftazidime, pefloxacin, augmentin, and Travid. *Klebsiella Pneumoniae* showed resistance to four antibiotics: Augmentin, Ceporex, Streptomycin and Ceftriaxone. Xu et al. [52] reported that *Klebsiella* strains isolated from poultry often carry extended-spectrum beta-lactamase (ESBL) genes such as blaCTX-M and blaSHV, which confer resistance to cephalosporins. *Salmonella* showed resistance to four antibiotics: Ciprofloxacin, Ceporex, Streptomycin and Ceftriaxone. This agrees with a previous study [43] that *Salmonella* is resistant to Ciprofloxacin. In their study, *Salmonella* isolated from poultry are resistant to antibiotics such as ampicillin, tetracycline, and ciprofloxacin. The presence of multidrug-resistant (MDR) *Salmonella* in poultry products raises concerns due to the potential transmission of these strains to humans [43].

The grilled chicken has a MAR range of 0.2 to 1.0. In the grilled chicken sample, the result revealed that *Proteus vulgaris* had the highest multiple antibiotic resistance index of 1.0 with a resistance pattern to all antibiotics studied: Ciprofloxacin, Ofloxacin, Augmentin, Pefloxacin, Ceftazidine, Gentamicin, Ciprofloxacin, Ceporex, Ceftriaxone and Streptomycin. *Bacillus subtilis* and *Salmonella enterica* had the lowest multiple antibiotic index. The MAR of raw chicken ranged between 0.5 and 1, while that of cooked chicken ranged between 0.4 and 1. Generally, it was observed that the pathogens showed high resistance to the following commonly used antibiotics: Augmentin, Streptomycin, Ceporex, ampiclox, and Ceftriaxone. In the grilled sample. Gram-negative isolates in this study indicated a high rate of resistance to Ceftriaxone and Streptomycin, and for Gram-positive isolates, it indicates a high rate of resistance to Ampiclox and Azithromycin. *Listeria* species was resistant to Ciprofloxacin, Azithromycin, Erythromycin, Gentamycin, Ampiclox, Amoxicillin and Rocephin, the study showed that *Listeria* is resistant to commonly used antibiotics this agrees with some previous reports [30] that while *Listeria* species is generally less resistant to antibiotics than other pathogens, there are emerging reports of resistance to commonly used antibiotics, including ampicillin and gentamicin, especially among strains isolated from poultry-processing environment. *Enterococcus* species are naturally present in the intestinal microbiota of poultry and can be transferred to meat during slaughter and processing [35]. Their ability to survive harsh conditions makes them persistent in the poultry supply chain. AMR in foodborne pathogens affects food safety standards, as industries must implement costly measures to control contamination and prevent transmission through the food chain [46].

5. Conclusion

This study underscores the critical need for coordinated efforts in monitoring and controlling AMR in the food chain. Reducing the use of antibiotics in poultry farming, practicing on-farm biosecurity, improving sanitation, vaccination, and better animal husbandry practices reduces the prevalence of foodborne pathogens. Improving hygiene during processing, and promoting safe handling and cooking practices are essential steps toward mitigating the public health threat posed by antimicrobial-resistant pathogens in food. There is a need to regulate the use of antibiotics in poultry farming and research on alternative approaches to antibiotic use.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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