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MICROBIAL ASSESSMENT OF READY-TO-EAT SHAWARMA OBTAINED FROM LAGELU LOCAL GOVERNMENT AREA, OYO STATE, NIGERIA

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Abstract: The consumption of ready-to-eat (RTE) foods has increased in recent years, especially in developing countries like Nigeria as population increases and demand for fast food increase. Despite the growing popularity of shawarma as a ready-to-eat food in Nigeria, particularly in semi-urban areas such as Lagelu Local Government Area (LGA) in Oyo State, there is a noticeable lack of empirical data on its microbial quality within this locality and the city of Ibadan in general. This study aimed to assess the microbial quality of ready-to-eat shawarma sold in selected locations within Lagelu Local Government Area, Oyo State, Nigeria, by identifying and isolating *Salmonella spp.*, *Shigella spp.*, and *Escherichia coli* from ready-to-eat shawarma sold by various outlets and vendors in Lagelu LGA of Oyo state. Shawarma sampled within Lagelu Government Area (Monatan, Mokola, Akobo, Ojoo, and Bodija) harbored *Salmonella spp.*, *Shigella spp.*, and *Escherichia coli* at different percentages with total viable counts of samples ranging from 5.2×10^4 to 3.10×10^5 CFU/g. *Shigella spp*. were the most frequently isolated organisms across all sampling locations, particularly in Ojoo and Bodija. This study highlights the need to improve hygiene practices in the preparation and handling of shawarma within the Lagelu Local Government Area. These findings emphasize the importance of a coordinated, multi-sectoral approach to food safety governance.

Key words: Shawarma, Ibadan, Shigella, Salmonella, Lagelu

1.0 Background of the Study

The consumption of ready-to-eat (RTE) foods has increased in recent years, especially in developing countries like Nigeria as population increases and demand for fast food increase. This surge can also be attributed to rapid urbanization, busy lifestyles, and the growing demand for convenience in food choices (Abalkhail, 2023). Ready-

to-eat foods do not require further preparation before consumption after being purchased and are popular among students, workers, and individuals with limited time for home-cooked meals (Al-Busaidi, et al., 2023).

Among the various types of RTE foods, shawarma has gained popularity in both urban and semi-urban areas of Nigeria. Originally, shawarma is a Middle Eastern delicacy, but it is now widely embraced and localized in Nigeria (Moawad and El-Saidi, 2019). It is made from grilled meat (often chicken or beef) wrapped in flatbread along with vegetables, sauces, and condiments. The cost is relatively low, and it has a rich flavor (Morshdy, et al., 2021).

However, the increased patronage of RTE foods such as shawarma has raised public health concerns. The risk of foodborne illnesses resulting from microbial contamination is one of the major issues. The preparation and handling of shawarma sometimes occurs in shops and environments that lack basic sanitation (El-Fakhrany et al., 2019). There are also problems with inadequate personal hygiene among food handlers, poor storage practices, and exposure of food to contaminated surfaces or utensils. These conditions create an environment favorable for the growth and transmission of pathogenic microorganisms such as *Escherichia coli*, *Salmonella* spp., *Staphylococcus aureus*, and *Listeria monocytogenes (*Ahmed et al., 2015*).*

This study aimed to assess the microbial quality of ready-to-eat shawarma sold in selected locations within Lagelu Local Government Area, Oyo State, Nigeria, by identifying and isolating *Salmonella spp., Shigella spp.,* and *Escherichia coli* from ready-to-eat shawarma sold by various outlets and vendors in Lagelu LGA of Oyo state. Lagelu was chosen due to being one of the largest local government areas in Oyo State, both in terms of population and landmass.

2.0 Materials and methods

Shawarma samples were purchased from vendors in the Monatan, Mokola, Akobo, Ojo and bodies areas all within Lagelu local government area of Ibadan. A sampling plan was developed using convenience sampling to obtain samples from vendors selling at the most popular relaxation spots within the local government area. Five shawarma samples were obtained.

Samples were put in sterile zip-lock plastic bags and transported in ice packs to the laboratory. General purpose as well as selective culture media, including Salmonella-Shigella agar, Macconkey agar, Nutrient agar, and Eosin methylene blue (EMB) agar**,** and Blood agar, were used to culture and isolate organisms, after which biochemical tests and microscopy were employed in identity the isolate. Antibiotic susceptibility tests were performed on the isolated bacteria using disc diffusion methods to determine the susceptibility or resistance of the isolate to a panel of antibiotics. The results for each isolate were documented.

2.1 Isolation and Identification of *Escherichia coli*, *Salmonella*, and *Shigella*

Approximately 25 grams of the shawarma sample was collected aseptically and then separated into shawarma bread, chicken, and salad. The shawarma bread, chicken, and salad were then placed into a sterile stomacher bag or blender jar. To this, 225 milliliters of sterile Buffered Peptone Water (BPW) was added. The mixture is then homogenized using a laboratory blender. Following homogenization, the samples were incubated at 37°C for 18 to 24 hours. For Salmonella isolation, a portion of the sample was inoculated into selective media and incubated at 37°C for 24 hours. For *Shigella* isolation, aliquots from each broth culture are streaked onto various selective and differential media. MacConkey agar is used to assess lactose fermentation, where *E. coli* appeared as pink colonies, whereas *Shigella* and *Salmonella* formed colorless colonies. Salmonella- shigella agar is used for

isolating *Salmonella* (which forms red colonies with black centers due to H_2S production) and *Shigella* which is an organism that produces red colonies without black centers.

Eosin Methylene Blue (EMB) agar helps in identifying *E. coli* by its characteristic green metallic sheen. Plates are incubated at 37°C for 24 hours. After incubation, colonies were observed and selected based on their morphological characteristics. Suspected colonies are subcultured on nutrient agar plates to obtain pure isolates. These pure cultures were then subjected to a series of biochemical tests to confirm their identity. The biochemical tests included the IMViC series urease test, triple sugar iron (TSI) slants for sugar fermentation, motility test, oxidase test and H_2S production. To preserve and maintain isolated organisms for extended periods, they were stored in agar slants. This method provides a stable environment that supports culture viability while minimizing the risk of contamination and genetic changes. The Gram staining technique was also employed to differentiate bacterial isolates into Gram-positive or Gram-negative groups based on their morphology.

2.2 Total Viable Count (TVC) of Shawarma Samples

Approximately 25 grams of the shawarma sample was aseptically weighed using a sterile spatula and transferred into a sterile stomacher bag containing 225 mL of sterile buffered peptone water or physiological saline. This yields a 1:10 dilution (10^{-1}). The sample was then homogenized using a stomacher or manually massaged for about 2–5 minutes to ensure an even distribution of microorganisms in the diluent. From this homogenized sample, serial dilutions are prepared by transferring 1 mL of the homogenate into 9 mL of sterile diluent in sterile test tubes to achieve 10^{-2} and 10^{-4} dilutions, depending on the expected microbial load. Using a sterile pipette, 0.1 mL of each dilution was plated onto sterile Nutrient Agar using the spread plate method. The inoculum was evenly spread over the agar surface using a sterile glass spreader or hockey stick.

After plating, the plates were incubated at 35°C–37°C for 24–48 hours in an incubator. Following incubation, the plates were selected for counting. Colonies are counted manually or using a colony counter. The TVC was calculated using the following formula: TVC (CFU/g) =Number of colonies× Dilution factor (ML) / Volume Plate

2.3 Bacterial antibiotic susceptibility assays and hemolysis test

The test for antimicrobial resistance and susceptibility were performed using the Kirby–Bauer disk diffusion technique on Mueller–Hinton agar plates. Antimicrobial disks impregnated with antimicrobials were placed on agar plates after 10 minutes and pressed to ensure contact. Plates were incubated at 37^{o}C for 24 hours. The diameters of the inhibition zones were measured using a ruler and recorded in millimeters on the under surface of the plate without opening the lid. Hemolysis test is an indicator of pathogenic potential. This test was conducted using blood agar, a nutrient-rich medium supplemented with approximately 5% defibrinated sheep or horse blood. To begin, sterile blood agar plates were prepared under aseptic conditions. A sterile inoculating loop is used to isolate the pure culture of the test organism. The plate was then incubated at 37°C for 24 to 48 hours, usually in a CO_2-enriched or candle jar environment to support the growth of fastidious organisms. After incubation, hemolysis zones surrounding the bacterial colonies were observed. Hemolysis is classified into beta, alpha, and gamma hemolysis.

2.4 Molecular Identification of the Isolates

The molecular identification of the bacterial isolates was carried out using 16S rRNA gene sequencing. Genomic DNA was extracted from each isolate using the boiling method. A loopful of a pure colony was suspended in

sterile distilled water and boiled at 95°C for 10 minutes. The lysate was then centrifuged to remove cell debris, and the DNA-containing supernatant was collected and stored at −20°C until further analysis.

Polymerase Chain Reaction (PCR) was performed to amplify the 16S rRNA gene using 27F and 1492R universal bacterial primers 27F and 1492R. The PCR reaction mixture contained the extracted DNA, primers, Taq polymerase, nucleotides, and buffer solution. The PCR conditions included an initial denaturation at 95°C for 5 minutes, followed by 35 cycles of denaturation at 95°C for 30 seconds, annealing at 55°C for 30 seconds, and extension at 72°C for 1 minute. A final extension was performed at 72°C for 5 minutes.

The confirmation of the amplified products was done by electrophoresis on a 1.5% agarose gel stained with ethidium bromide. Bands of approximately 1500 base pairs were visualized under UV light, indicating the successful amplification of the 16S rRNA gene. The PCR products were purified and sent for Sanger sequencing. The obtained sequences were analyzed using the Basic Local Alignment Search Tool (BLAST) on the NCBI GenBank database. The identity of each isolate was determined by comparing the sequences with known bacterial sequences in the database. Isolates with ≥98% similarity were confirmed as *Shigella*, *Salmonella,* and *E. coli*.

3.0 Results

Table 1: Total Viable count of Shawarma Samples

Sample Code	Location	Dilution Factor	Colony Count (CFU)	TVC (CFU/g)
S1	Monatan	10^{-2}	152	1.52×10^5
S2	Mokola	10^{-2}	278	2.78×10^5
S3	Akobo	10^{-2}	95	9.5×10^4
S4	Ojo	10^{-2}	310	3.10×10^5
S5	Bodija	10^{-2}	180	1.80×10^5
S6	Monatan	10^{-4}	80	8.0×10^5
S7	Mokola	10^{-4}	136	1.36×10^6
S8	Akobo	10^{-4}	47	4.7×10^5
S9	Ojo	10^{-4}	183	1.83×10^6
S10	Bodija	10^{-4}	100	1.00×10^6

Table 2 Biochemical results, Gram staining, and morphology of isolates obtained in the Monatan

Isolate Code	Indole Test	Methyl red Test	VP Test	Citrate Test	Urease Test	TSI Test	H2S Test	Gram staining/morphology	Suspected Identity
MnB 10-2	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
MnB 10-4	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
MnC1 10-2	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*

MnC2 10-4	_	+	_	+	_	+	+	Gram-negative rod	*Salmonella sp.*
MnC3 10-4	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
MnS 10-2	_	+	_	+	_	+	+	Gram-negative rod	*Salmonella sp.*
Mns 10-4	+	+	_	_	_	+	_	Gram-negative rod	*Escherichia coli*
MnC4 10-2	+	+	_	_	_	+	_	Gram-negative rod	*Escherichia coli*
MnC5 10-4	+	+	_	_	_	+	_	Gram-negative rod	*Escherichia coli*

Table .3 Biochemical results, Gram staining, and morphology of isolates obtained from Mokola and Akobo

Isolate Code	**Indole Test**	**Methyl red Test**	**VP Test**	**Citrate Test**	**Urease Test**	**TSI Test**	**H2S Test**	**Gram staining/morphology**	**Suspected Identity**
OJC1 10-4	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
OJC2 10-2	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
OJB1 10-2	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
OJB2 10-4	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
OJS1 10-2	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
OJC3 10-2	_	+	_	+	_	+	+	Gram-negative rod	*Salmonella sp*
OJC4 10-4	_	+	_	+	_	+	+	Gram-negative rod	*Salmonella sp*
OJS2 10-4	+	+	_	_	_	+	_	Gram-negative rod	*Escherichia coli*
OJB3 10-4	+	+	_	_	_	+	_	Gram-negative rod	*Escherichia coli*

Table 4. Biochemical results, Gram staining, and morphology of isolates obtained from Ojoo

Isolate Code	Indole Test	Methyl red Test	VP Test	Citrate Test	Urease Test	TSI Test	H2S Test	Gram staining/morphology	Suspected Identity
MKC1 10-4	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
MKC2 10-2	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
MKC3 10-4	_	+	_	+	_	+	+	Gram-negative rod	*Salmonella sp.*
MKS1 10-4	+	+	_	_	_	+	_	Gram-negative rod	*Escherichia coli*
MKS2 10-4	+	+	_	_	_	+	_	Gram-negative rod	*Escherichia coli*
AKS1 10-4	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
AKB1 10-4	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
AKC1 10-4	_	+	_	+	_	+	+	Gram-negative rod	*Salmonella sp*

Table 5: Biochemical results, Gram staining, and morphology of isolates obtained from Bodija

Isolate Code	Indole Test	Methyl red Test	VP Test	Citrate Test	Urease Test	TSI Test	H2S Test	Gram staining/morphology	Suspected Identity
MKC1 10-4	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
MKC2 10-2	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
MKC3 10-4	_	+	_	+	_	+	+	Gram-negative rod	*Salmonella sp.*
MKS1 10-4	+	+	_	_	_	+	_	Gram-negative rod	*Escherichia coli*
MKS2 10-4	+	+	_	_	_	+	_	Gram-negative rod	*Escherichia coli*
AKS1 10-4	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
AKB1 10-4	+	+	_	_	_	_	_	Gram-negative rods	*Shigella Sp.*
AKC1 10-4	_	+	_	+	_	+	+	Gram-negative rod	*Salmonella sp*

Table 6: Antibiotic susceptibility test and inhibition zone in diameter (mm) of bacteria isolate from Monatan

S/N	Isolates	CH	SP	CPX	AM	AU	CN	PEF	OFX	S	SXT
1	MnB 10-2	14	18	16	6	6	6	16	7	6	6
2	MnB 10-4	10	6	12	6	16	6	14	6	6	6
3	MnC1 10-2	6	11	14	6	6	6	6	7	6	6
4	MnC2 10-4	6	6	27	14	6	12	16	18	14	10
5	MnC3 10-4	6	6	19	6	8	6	6	20	6	6
6	MnS 10-2	6	6	20	6	6	6	6	11	6	6
7	Mns 10-4	6	6	19	6	6	6	6	6	6	6
8	MnC4 10-2	6	7	17	6	6	6	6	20	6	12
9	MnC5 10-4	6	12	19	6	6	7	6	12	6	6
10	MnB 10-2	6	6	17	10	6	6	10	20	10	6
11	MnB 10-4	6	6	17	9	14	6	12	19	6	6
12	MnC1 10-2	6	12	19	6	6	6	6	12	9	10
13	MnC2 10-4	6	6	21	12	6	6	6	17	6	6
14	MnC3 10-4	11	6	20	6	6	6	14	25	15	6

NOTE: PEF= PEFLOXACIN 30ug, CN= GENTAMYCIN 10ug, AM= AMOXACILIN 30ug, CPX= CIPROFLOXACIN 10ug,S= STREPOTOMYCIN 30ug, SXT= SEPTRIN 30ug, CH= CHLORANPHENICOL 30ug,AU = AUGMENTIN 30ug,OFX= TARIVID 10ug, SP=SPARFLOXACIN 10ug.

Table 7: Antibiotic susceptibility test and inhibition zone in diameter (mm) of bacteria isolate from Mokola and Akobo

S/N	ISOLATE CODE	CH	SP	CPX	AM	AU	CN	PEF	OFX	S	SXT
1	MKC1 10-4	14	18	16	6	6	6	16	7	6	6
2	MKC2 10-2	10	6	12	6	16	6	14	6	6	6
3	MKC3 10-4	6	11	14	6	6	6	6	7	6	6
4	MKS1 10-4	6	6	27	14	6	12	16	18	14	10
5	MKS2 10-4	6	6	19	6	8	6	6	20	6	6
6	AKS1 10-4	6	6	20	6	6	6	6	11	6	6
7	AKB1 10-4	6	6	19	6	6	6	6	6	6	6
8	AKC1 10-4	6	7	17	6	6	6	6	20	6	12

Table 8: Antibiotic susceptibility test and inhibition zone in diameter (mm) of bacteria isolate from Ojo

S/N	Isolates	CH	SP	CPX	AM	AU	CN	PEF	OFX	S	SXT
1	OJC1 10-4	14	18	16	6	6	6	16	7	6	6
2	OJC2 10-2	10	6	12	6	16	6	14	6	6	6

3	OJB1 10-2	6	11	14	6	6	6	6	7	6	6
4	OJB2 10-4	6	6	27	14	6	12	16	18	14	10
5	OJS1 10-2	6	6	19	6	8	6	6	20	6	6
6	OJC3 10-2	6	6	20	6	6	6	6	11	6	6
7	OJC4 10-4	6	6	19	6	6	6	6	6	6	6
8	OJS2 10-4	6	7	17	6	6	6	6	20	6	12
9	OJB3 10-4	6	12	19	6	6	7	6	12	6	6

NOTE: PEF= PEFLOXACIN 30ug, CN= GENTAMYCIN 10ug, AM= AMOXACILIN 30ug, CPX= CIPROFLOXACIN 10ug,S= STREPOTOMYCIN 30ug, SXT= SEPTRIN 30ug, CH= CHLORANPHENICOL 30ug,AU = AUGMENTIN 30ug,OFX= TARIVID 10ug, SP=SPARFLOXACIN 10ug.

Table 9: Antibiotic susceptibility test and inhibition zone in diameter (mm) of bacteria isolate from Monatan

S/N	Isolates	CH	SP	CPX	AM	AU	CN	PEF	OFX	S	SXT
1	BOC1 10-4	14	18	16	6	6	6	16	7	6	6
2	BOB1 10-2	10	6	12	6	16	6	14	6	6	6
3	BOC2 10-2	6	11	14	6	6	6	6	7	6	6
4	BOB2 10-4	6	6	27	14	6	12	16	18	14	10
5	BOB3 10-4	6	6	19	6	8	6	6	20	6	6
6	BOC3 10-2	6	6	20	6	6	6	6	11	6	6

NOTE: PEF= PEFLOXACIN 30ug, CN= GENTAMYCIN 10ug, AM= AMOXACILIN 30ug, CPX= CIPROFLOXACIN 10ug, S= STREPOTOMYCIN 30ug, SXT= SEPTRIN 30ug, CH= CHLORANPHENICOL 30ug,AU = AUGMENTIN 30ug,OFX= TARIVID 10ug, SP=SPARFLOXACIN 10ug.

Table 10: Hemolysis Test for isolates from Monatan, Mokola , Akobo, and Bodija

Isolate	Beta (Complete Hemolysis)	Alpha (Incomplete Hemolysis)	Gamma (Non-Hemolysis)
MnB 10-2	+	-	-
MnB 10-4	+	-	-
MnC1 10-2	+	-	-
MnC210-4	+	-	-
MnC3 10-4	+	-	-
MnS 10-2	+	-	-
Mns 10-4	+	-	-
MnC4 10-2	-	+	-
MnC5 10-4	-	+	-
MKC1 10-4	+	-	-
MKC2 10-2	+	-	-

MKC3 10-4	+	-	-
MKS1 10-4	+	-	-
MKS2 10-4	+	-	-
AKS1 10-4	+	-	-
AKB1 10-4	+	-	-
AKC1 10-4	-	+	-
BOC1 10-4	-	+	-
BOB1 10-2	+	-	-
BOC2 10-2	+	-	-
BOB2 10-4	-	+	-
BOB3 10-4	-	+	-
BOC3 10-2	+	-	-

Table 11: Hemolysis test results for isolates from Ojoo

Isolate	Beta (Complete Hemolysis)	Alpha (Incomplete Hemolysis)	Gamma (Non-Hemolysis)
OJC1 10-4	+	-	-
OJC2 10-2	-	+	-
OJB1 10-2	-	+	-
OJB2 10-4	+	-	-
OJS1 10-2	+	-	-
OJC3 10-2	-	+	-
OJC4 10-4	-	+	-
OJS2 10-4	+	-	-
OJB3 10-4	-	+	-

Molecular Identification of the Species

Three isolates were selected on the basis of the isolates with the highest antimicrobial resistance and hemolysis. They were taken for molecular analysis, and this is the result of the analysis.

Isolate 1: MnC3 10-4- ***Shigella Sp.***

GGGGGGMRAACTTAACGCGTTAGCTCCGGAAGYCACGSCTCAAGGGCACAACCTCCAAGTCGAC
ATCGKTTACGGSGKGGACTACCAGGGKATCTAATCCTGKTTGCTCCCCACGCTTTCGCACCTGARC
GKCAGTCTTTGTCCAGGGGGCCGSCTTCSCCACCGGKATTCCTCCAGATCTCTACSCATTTCACCGS
TACACCTGGAATTCTACCCCCCTYTWCAAGACTCTAGCCTGCCAGTTTYSAATGSAGTTCCCAGGK
TGAGCCCGGGGATTTCACATCCGAMTTGACARAMCGCCTGSGKGCGCTTTACSCCCARWAATTCC
SATTAACGCTTGSACCCTYCGKATTACCGSGGSTGSTGGSACGGARTTARCCGGTGSTTYTTCTGYG
GGKAACSKCAATYGCTGAGGTTATTAACCACRACGCCTTCCTYCCCGCTGAAAGTACTTTACAAM

CCSAARGSCTTCTTCATACACSCGGSATGGSTGCATYAGGSTTGCGCCCATTGKGCAAWAWTCCCC
AMTGSTGSCTYCCCKARGARTCTGGACCGTGTCTYARTTCCAGTGTGGSTGGWCATCCTCTCAGAM
CAGCTARGGAWCGTCGCCTAGGTGARCCGTTACCCCACCTACTAGCTAATYCCATCTGGGCACAT
CTGATGGCAAGAGGCCCGAAGGKCCCCCTCTTTGGTCTTGCGACGKTRWGCGGTATTAGCTACCG
TTTCCASTAGTTATCCCCCTCCATCAGGCAGTWTCCSRGASRTTACTCACCCGTCCGTCACTCGTCA
CCCGAGARCAARCTCTCWGTGCTAYCGTTCKACTTGCATGTGWTTGCCTGCCGCCAGCGTTCAAT
CTGAGYCAGGTTCAAAACTMTKTWAAA.

Isolate 2: Mns 10-4 ***Escherichia coli strain***

GSGGGGTCGRACTTAWCGGCGTTAGCTGCGSCACTMAGATCTCAAGGRTMCMAACKGCTAGTCG
ACATCGTTTACGGCGTGGACTACCAGGGTATCTAATCCTGTTTGCTCCCCACGCTTTCGCACCTSAG
YGTCAGTATYWGTCCAGGKGGYCGCCTTCGCCACYGGTGTTCCTYCCTATMTCTACGCATTTCAC
CGCTACACMKGRAATTCYACCMCCCTCTACCGTACTCTAGCTTGGYAGTTTYGGATGCAGTTCCC
AGGTTGAGCCCGGGGATTTCACATCCAACTTGMYRAACCRCCTRCGCGCGCTTTACGCCCAGTAA
TTCCGATTAACGCTTGCACCCTYCGTATTACCGCGGCTGCTGGCACGRAGTTAGCCGGTGCTTMTT
CTGYTRGTAACGTCAAAACASMAAGGTATTAACTTACTGSCCTTCCTCCCAACTTAAAGTGCTTTA
CAATCCGAAGACCTTCTTCAYACACGCGGCATGGCTGGATCAGGCTTKCGCCCATTGTSCAATATT
CCCCACTGCTGCCTCCCGTAGGAGTCTGGACCGTGTCTCAGTTCCAGTGTGACTGATCATCCTCTC
AGACCAGYTACGGATCGTCGCCTWGGTRRGCCTTTACCCCACCAACTAGCTAATCCSAYCTAGGC
TCATCTGATRGCGTGAGGTCCGAAGATCCCCCACTTTCTCCCTSMGGACGTATGCGGTATTAGCKM
CCGTTTCCRGACGTTATCCCCCWCTAYCAGGCAGATTCCTAGGCATTACTCACCCGTCCGCCGCTG
AATCCASGAGMAAGMTCCCTTCRTCCGCTCGAYTTGCATGTGTGAGGYCTGCCGSCMGCGWTCAA
TCTGAGCCAGGATCRATCTCTATCTAA

Isolate 3: AKB1 10-4 ***Shigella Sp.***

GGGGGGARGAMTTAWCGCGTTAGCTGCGCCACTGARAGTAAASGGYMCCCAMGRTAARTYSATT
TYKTTWAGGGSGGGAACTACCAGGGWATCTAATCGGGTTGGTTCCCCACTTTTTCSWGCCTCAGG
GCAAAAGYGGGCCCAGGGAGCTGCCTTCGCTGGGATTCCCCTCCTGATCTCTAATTTTTRACTGTT
ACACCAAATTCCRATCACCTTTCTAGATTTYTAGTCRCCYATTTSGGGGMAGTTGGGGTGAGSCCC
GGGGGTTTTCCAMCAMTTAGAAACCCCCCCCTACGCACTTTTTACCCCCRRAAATCCCAAKTAACT
TTKGCCCCTTTYATTTTACCGSGTGTGGTGGMMAAAATTWACCCGGSTTTTTTTCTTTGGGWACCS
ATATMCTAACCGGGAATAASYYATTTGSATTTCTTCCCACTTAAASGGGTTTWCCAACCGGAAGGC
TTTCTTTMCCCCCCRGKGAGGGTTATTCCAGGCTTGCGCCCTTTGTCMAAAATTCCCCCKTGTTGC
CYCCCCARGGAGGCTGAACAGSSTCTCAGACCAAGTGTGGWTGATCATCTTSTMAAACCATTACC
GSACCAACTCTTTGATGGGCCTTTMCCCCACCAATTASCTATWCAAACTTCGCCTCAYTSAATCGS
GCAGGACCCGAAGGCCCCCTGMTTCCACACGTRGGGGRAWGGSGGAATTAGCSWARGTTTWCCG
TACATTAATCCCCCACSACARAGYAAATTCCKAAKGWATTCCTYAMCCCRTCCCCCACTCGACAC
CCGAAGGAGCASCCTMYCTYTGYGCTSCTATATCAKTTGTATGGGSTTKCGCCYASCAGCCAATAT
TAACTCTTGKCCAAGTTCMTGACTMT

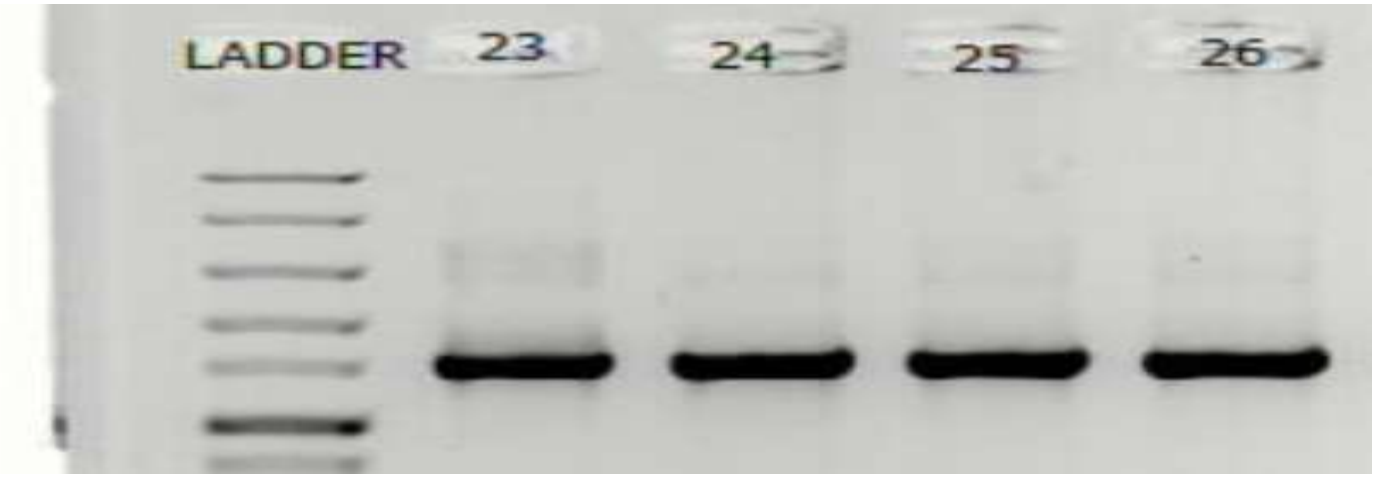

Figure 4.1: Visualization of DNA bands on Gel Eletrophoresis

Discussion

Ready-to-eat (RTE) foods, including shawarma, have become increasingly popular in urban and peri-urban areas due to their convenience, affordability, and widespread availability (Siddique, et al., 2024). Shawarma, a meat-based RTE delicacy, is commonly prepared and available in Nigerian outlets. However, its popularity has raised considerable public health concerns regarding hygiene.

Studies have shown microbial contamination of shawarma and other RTE foods are contaminated by bacteria across different countries. Raza et al. (2021) and Makinde et al. (2023) reported the isolation of *Salmonella spp.* and *Shigella spp.* from street-vended foods in Pakistan and Lagos, Nigeria, respectively, with total viable counts exceeding permissible levels. Similarly, Al-Busaidi et al. (2023) found that many RTE sandwiches in Oman were microbiologically unsafe because of post-processing contamination and poor food handling [1].

Salmonella sp. Escherichia coli and *Shihella sp.* have been detected in shawarma across various regions, including Egypt (Ahmed et al., 2015; Salem et al., 2015), Jordan (Nasraween et al., 2022), Zambia (Chileshe et al., 2024), and Nigeria (Odu & Akano, 2012), indicating systemic lapses in food safety practices. These pathogens are typically introduced through contaminated raw materials, undercooked meat, unclean utensils, or unhygienic food handling.

The rise in antibiotic resistance among bacterial isolates from RTE foods presents a dual challenge of foodborne illness and treatment failure. Mohamed and El-Zahaby (2024) demonstrated that bacteria from shawarma not only secreted harmful toxins but also exhibited multi-drug resistance [11]. This finding was corroborated by Sotohy Sotohy and Abd EL-Malek, (2019), who identified methicillin- and vancomycin-resistant *Staphylococcus aureus* in RTE meats, underscoring the global emergence of antimicrobial-resistant foodborne pathogens.

Despite the growing body of literature on microbial contamination of shawarma in major urban centers, there is limited data specific to urban areas such as Lagelu in Oyo State, Nigeria. Understanding the microbial quality of shawarma sold in such locations is essential for risk assessment and public health planning. Therefore, this study aimed to assess the microbial load, isolate pathogenic bacteria, evaluate their antibiotic resistance profiles, and determine the hemolytic activity of isolates from shawarma samples collected in selected locations within Lagelu LGA. The study builds upon global and regional findings (e.g., Saeed & Mohammad, 2021; Ahmed et al., 2015; Nasraween et al., 2022) while addressing a local knowledge gap in Nigeria that is critical for food safety policy formulation.

Shawarma sampled within Lagelu Government Area (Monatan, Mokola, Akobo, Ojoo, and Bodija) harbored diverse microbial contaminants, with total viable counts (TVCs) of 5.2×10^4 to 3.10×10^5 CFU/g. These values exceed the acceptable limits recommended for these sorts of foods, making the food potential public health risk. Similar high microbial loads have been reported by Al-Busaidi et al. (2023), who found that RTE sandwiches often exceed safety thresholds due to post-processing contamination and poor handling [1].

Isolation of *Escherichia coli*, *Salmonella spp.,* and *Shigella spp. from* shawarma is particularly concerning. These pathogens are commonly associated with foodborne diseases and indicate fecal contamination, poor hygiene practices, or cross-contamination during food preparation. Results from this study are in tandem with the findings of Raza et al. (2021), who reported Salmonella *spp.* to be a prominent bacteria in fast foods in Pakistan and emphasized the implications for consumer health [2].

Shigella spp. were the most frequently isolated organisms across all sampling locations, particularly in Ojoo and Bodija. Its frequent presence aligns with earlier studies by Odu and Akano (2012) and Makinde et al. (2023), who reported Shigella as a dominant pathogen in shawarma sold in Nigeria. This indicates persistent hygiene issues in RTE food preparation environments, which are often intensified by inadequate sanitation facilities and improper food handling.

Salmonella spp. were found in vendors sampled in Mokola and Akobo, which agrees with the findings of Ahmed et al. (2015) and Salem et al. (2015), who observed Salmonella contamination in beef and chicken shawarma in Egypt. Cross-contamination from raw ingredients or undercooked meat may explain this trend, which is consistent with the reports of Nasraween et al. (2022) on microbial load linked to poor food handler compliance with safety practices.

E. coli was isolated predominantly from the Monatan and Ojoo samples, indicating contamination likely due to unhygienic conditions, unwashed vegetables, or inadequate cooking temperatures. Chileshe et al. (2024) showed in their study in Zambia and Egypt that *E. coli* was a major contaminant present in shawarma, and their presence might be due to improper cooking and post-cooking handling.

The antibiotic susceptibility tests revealed worrying resistance patterns among the isolates. For instance, many isolates displayed resistance to amoxicillin (AM), augmentin (AU), gentamicin (CN), and streptomycin (S). This antimicrobial resistance (AMR) is consistent with studies by Mohamed and El-Zahaby (2024), who noted that bacteria isolated from shawarma demonstrated multi-drug resistance (MDR), posing a significant threat to treatment efficacy. The ability of these organisms to resist multiple antibiotics, especially ciprofloxacin and pefloxacin, may indicate the overuse or misuse of antibiotics in livestock and clinical settings. The hemolysis test results also revealed that many isolates demonstrated beta-hemolysis, which is associated with the presence of hemolysins, which are virulence factors that contribute to the pathogenicity of bacteria. This further supports the assertion by Mohamed and El-Zahaby (2024) that many toxin-producing bacteria are present in shawarma and may pose severe health risks to consumers if ingestion occurs.

Interestingly, there were location-based variations in microbial counts and species distribution. For example, Ojoo had the highest TVC and most diverse pathogen profile, possibly due to high foot traffic, street vending, and poor enforcement of food safety regulations. This observation mirrors the findings of Saeed and Mohammad (2021), who found that shawarma sold in more congested or informal settings had higher contamination rates.

The role of vendors and their hygiene practice cannot be ignored in the quest to reduce microbial load and contamination in food sold on the streets or in restaurants cannot be ignored. Nasraween et al. (2022) reported

poor basic hygiene practices among handlers, with problems identified as improper glove use, lack of handwashing, and inadequate storage conditions, all of which were likely mirrored in the current study area.

The findings clearly demonstrate that shawarma sold in the Lagelu Local Government Area is microbiologically unsafe and poses significant health risks to consumers. This is particularly concerning given the detection of enteric pathogens and antimicrobial-resistant bacteria in the analyzed samples. These results highlight the urgent need for public health interventions. The key recommendations include improving food safety training for vendors, conducting regular health inspections and microbial monitoring, increasing public awareness about the potential risks associated with ready-to-eat foods, and strictly enforcing food hygiene regulatory standards.

Conclusions and recommendations

This study highlights the critical need to improve hygiene practices in the preparation and handling of shawarma within the Lagelu Local Government Area. The presence of harmful bacteria such as *Shigella*, *Salmonella*, and *Escherichia coli*, along with the high prevalence of antibiotic resistance, reflects patterns commonly observed in many developing countries. These findings emphasize the importance of a coordinated, multisectoral approach to food safety governance. In line with similar research conducted in Nigeria, Egypt, Jordan, Zambia, and Pakistan, the study reinforces the global challenge of ensuring the microbiological safety of ready-to-eat (RTE) foods. In other to address these concerns, the study recommends improved hygiene practices among food vendors, routine surveillance and monitoring by public health authorities, and the implementation of larger-scale studies in future research efforts to better understand and mitigate these risks.

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