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Prevalence and antibiogram of *Salmonella species* isolated from marketed pork meats in Port Harcourt Metropolis

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Abstract
One of the major food-borne pathogens that causes gastroenteritis and typhoid is the species of *Salmonella*. This study investigated the prevalence and antibiogram of *Salmonella species* in marketed pork meat in Port Harcourt. One hundred and twenty (120) samples comprising fresh pork, roasted pork, wastewater, knife and table swabs were collected from different vendors in the Port Harcourt metropolis. Standard microbiological techniques were employed to isolate and identify *Salmonella* strains, and the susceptibility pattern were determined using disc diffusion methods. Total heterotrophic bacterial (THB), total *Salmonella, Shigella,* coliform and staphylococcal load of the pork ranged from 1.05±0.33 to 1.51±0.49×10^7, 3.7±0.46 to 7.4±0.89×10^4, 0.85±0.11 to 1.8±0.27×10^5, 2.0±1.12 to 7.8±5.1×10^5 and 1.3±0.5 to 3.3±0.6×10^5 CFU/g, respectively. Despite the observed differences in bacterial counts, there was no significant difference (p>0.05) in the *Salmonella* and THB of the pork samples. The THB, total *Salmonella*, *Shigella,* coliform and staphylococcal load of the butcher table ranged from 1.0±0.3 to 1.8±0.2×10^7, 3.6±0.5 to 8.1±0.9×10^4, 4.4±0.7 to 10.3±1.2×10^4, 2.2±0.5 to 4.2±2.2×10^5, and 1.3±0.8 to 2.4±0.6×10^5 CFU/m^2, respectively. The THB, total *Salmonella, Shigella,* coliform and staphylococcal load of the knives ranged from 40.91±0.4 to 1.5±0.4×10^7, 1.4±0.2 to 5.4±0.6×10^4, 2.7 ±0.3 to 8.1±0.8×10^4, 1.1±0.6 to 2.2±1.1×10^5 and 1.0±0.7 to 1.5±1.2×10^5 CFU/m^2, respectively, while the THB, total *Salmonella, Shigella,* coliform and staphylococcal load of the wastewater ranged from 0.99±0.28 to 1.3±0.7×10^7, 6.0±0.6 to 9.3±0.5×10^4, 7.6±0.8 to 14.3±1.6×10^4, 2.7±1.7 to 4.2±3.4×10^5 and 1.4±0.4 to 3.8±2.2×10^5 CFU/mL. No significant difference (p>0.05) was recorded in the *Salmonella* counts of all the wastewater samples, knives and Tables in all locations. Eighty-seven *Salmonella* isolates were identified. Results showed that pork meat samples from Rumuolumeni recorded the highest (20.7%) *Salmonella* prevalence while the least (5.7%) was from Iwofe locations. *Salmonella* spp from fresh pork samples displayed a higher virulence compared to those associated with the roasted pork samples. The multidrug resistance showed that 100% of the salmonella had a MAR index greater than 0.2. They were more susceptible to Levofloxacin (90.8%) and resistant to Amoxicilin Clavulanate (4.6%). The bacterial load in the pork meats were generally high with a high prevalence of *Salmonella* in both the fresh and roasted pork meat which can cause consumer diseases and act as vehicle for the transfer of antibiotic resistance gene in food. Thus, stringent measures are recommended in the processing and preparation of pork meat before consumption.

Keywords: *Salmonella,* Port Harcourt Metropolis, marketed pork

Introduction
Foodborne diseases are widespread and are of great public health concern in the modern world. In developing countries, the greater populace is largely affected by foodborne infections (Akbar and Anal, 2013) [2]. Foodborne disease apart from affecting the health and well-being of individuals equally affects the social and economic productivity of the countries. The main factors contributing to the increased burden of food-borne diseases, especially in Africa, are the people's poor hygiene practices. Poor personal hygiene among food handlers coupled with the inadequate handling of meat products in abattoirs could be possible sources of acquiring microbial pathogens that cause foodborne infections (Akbar and Anal, 2013) [2].

Pork is one of the most widely eaten meats in the world, accounting for about 38% of meat production worldwide, although consumption varies widely from place to place (Chaudhary *et al.*, 2015) [8]. According to the Food and Agriculture Organization of the United Nations, the world's pork production reached 114.2 million tons in 2012.

With their high nutritional value, pork and sausage products remain highly prized and it is considered one of the foods of choice because their nutritional value (rich in nutrients, including essential amino acids), is also well digestible, which justifies their rapid development in the world of the pig meat products industry and all related commercial transactions (Alfred *et al.*, 2019) [3]. Pork is prepared and consumed in different forms. In Nigeria, it is prepared grilled, boiled, or dried. In a previous study, pork is reportedly consumed in different forms: Braised, grilled or boiled with vegetables (soup), in places of relaxation, leisure and during celebrations (Alfred *et al.*, 2019) [3]. Pork consumption per capita has been increasing from 8 kg in 1990 to 30 kg in 2019.

Salmonella is one of the most common causes of foodborne diarrhoeal diseases worldwide and most of these infections are zoonotic and transmitted from apparently healthy carrier animals to humans through contaminated foods. The main reservoirs of zoonotic *Salmonella* are food animals, and the main sources of infections in industrialized countries are animal-derived products, notably fresh meat products, poultry, and eggs (Heredia and García, 2018) [15]. In developing countries, however, contaminated water, vegetables, and human-to-human transmission contribute to a comparatively larger proportion of human cases than those in developed countries (WHO, 2020) [20]. Microbial food safety is an increasing public health concern worldwide and the importance of food as a vehicle for the transmission of many diseases has been documented for a long time, especially in developing countries where hygienic standards are not strictly enforced and followed (Harakeh *et al.*, 2005) [14]. There is an upsurge of pork production in the state as people source alternate meat supplies or proteins. Thus, many individuals are into selling pork either fresh, grilled or boiled with little knowledge of hygiene. This study therefore investigated the prevalence of *Salmonella species* in pork meat sold in different parts of Port Harcourt, Rivers State.

Materials and Methods

Study Design

The study was a cross-sectional study which adopted a completely randomized sampling approach where each location had an equal chance and number of samples represented within the sampled frame. The study was conducted from July 2023 to November 2023.

Sample Size and Sample Collections

A total of one hundred and twenty (120) samples were obtained from eight sampling locations. The samples were fresh and roasted pork meat, swab samples from tables and knives, and water samples used in washing the pork meats from the different locations. The samples were collected in sterile containers and carried in ice-pack containers to the Microbiology Laboratory, Rivers State University for immediate analysis.

$$N = \frac{Z^2 (p \times q)}{d^2}$$

Equation 1

Where;
N=required sample size
P = prevalence of *Salmonella* sp. in Pork = 8% = 0.08
Z = 1.96 at 95% confidence limit
Q = 1-p = 0.92
d = margin of error = 5% at a confidence level of 95%

Microbiological Analysis

Preparation of Stock Samples

A Twenty-five (25g) gramms of cut pork was immersed in (225) ml Buffered Peptone Water (BPW) in a sterile conical flask to serve as stock for the fresh and roasted pork, while for the wastewater, 1mL (water from the washed pork) was diluted into 9mL normal saline which served as a stock for the wastewater. The swab samples from the knife and tables were immersed into sterile 9mL normal saline, respectively to obtain their stock. After the preparations of the different stocks, a ten-fold serial dilution was carried out using sterilized normal saline as the diluent. In this method, 1mL of the stock was withdrawn and transferred into a test tube containing sterile 9mL normal saline to give a dilution of 1:100. The dilution was repeated in a stepwise fashion by transferring 1mL from the 1:100 dilution to another test tube containing 9mL sterile normal saline. This was done until dilutions of 10^{-6} were obtained.

Bacterial Enumeration

Aliquots of (0.1ml) dilutions of the diluted samples (pork, wastewater, knives and tables) were plated out on well labelled freshly prepared pre-dried plates containing nutrient agar (NA) in duplicates for enumeration and isolation of total heterotrophic bacteria. Aliquots of 10^{-2} and 10^{-3} dilutions were plated on mannitol salt agar (MSA) for enumeration and isolation of total *Staphylococcus* sp, while aliquots of 10^{-1} and 10^{-2} dilutions were plated out on well-labelled freshly plated out on prepared pre-dried plates containing Eosin-methylene blue (EMB) agar, MacConkey Agar (MCA), *Salmonella-Shigella* agar and Thiosulfate Citrate Bile Salts Sucrose agar (TCBS) Plates for the enumeration and isolation of faecal coliform, total coliform, *Salmonella*, *Shigella* and *Vibrio* in the samples. All the inoculation were done in duplicates and incubated at 37°C for 24-48 hours. On observation of growth after incubation, colony counts were done and results were recorded.

Confirmation test for *Salmonella* spp.

The suspected colonies of *Salmonella* (colourless colonies with a black centre) from the pure culture were cultured on freshly sterile prepared nutrient agar for biochemical tests (e.g. sugar fermentation, citrate, indole, lysine, H_2S, and urease tests) and serology (Antiserum *Salmonella* Polyvalent-O).

Determination of Antibiogram of Isolates (Kirby Bauer Disc Diffusion)

A sterile swab stick was dipped into a test tube containing the bacterial suspension which has been standardized to 0.5 McFarland Turbidity Standard. The swab was evenly swabbed on the surface of the prepared Mueller Hinton agar plates. The agar was allowed to dry for about 3-5minutes. With Sterile forceps, the impregnated antibiotic discs were placed evenly on the surface of the inoculated plate and incubated in an inverted position aerobically at 37 ºC for 24 hours. After incubation, the test plates were examined for zones of inhibition. The diameter of each zone of inhibition was measured in mm using a ruler on the underside of the plate and recorded (CLSI, 2017) [9].

Determination of Multiple Antibiotic Resistance Index (MARI)

Multiple antibiotic resistance (MAR index was calculated using the formula MAR=a/b, where a stands for the number of antibiotics to which the test isolate showed resistance and b stands for the total number of antibiotics to which the test isolate was evaluated for susceptibility (Apun *et al.*, 2008) [5].

Statistics

Analysis of variance (ANOVA) was carried out to check for significant differences in the bacterial counts. The Duncan multiple range test was used in separating means and the significant level used was $p<0.05$ (95% confidence interval). All the analysis was carried out using the statistical package for social sciences (SPSS v27).

Results

The results of the bacterial load of Pork meat samples in Table 1 showed that the mean total heterotrophic bacterial load, total *Salmonella*, *Shigella*, coliform and staphylococcal load were 1.05±0.33 to 1.51±0.49×10^7, 3.7±0.46 to 7.4±0.89×10^4, 0.85±0.11 to 1.8±0.27×10^5, 2.0±1.12 to 7.8±5.1×10^5 and 1.3±0.5 to 3.3±0.6×10^5 CFU/g, respectively.

The results of the bacterial load of Tables used for the slaughter of Pork Meat in all locations in Table 2 showed that the mean total heterotrophic bacterial load, total *Salmonella*, *Shigella*, coliform and staphylococcal load was 1.0±0.3 to 1.8±0.2×10^7, 3.6±0.5 to 8.1±0.9×10^4, 4.4±0.7 to 10.3±1.2×10^4, 2.2±0.5 to 4.2±2.2×10^5, and 1.3±0.8 to 2.4±0.6×10^5 CFU/m^2, respectively. Results further showed that despite the high *Salmonella* load recorded in the samples from the Rumuokoro location, it was not significantly different ($p>0.05$) from the *Salmonella* counts in other locations.

The results of the bacterial load of the knife used for cutting the pork meat in Table 3 showed that the mean total heterotrophic bacterial load, total *Salmonella*, *Shigella*, coliform and staphylococcal load was 0.91±0.4 to 1.5±0.4×10^7, 1.4±0.2 to 5.4±0.6×10^4, 2.7±0.3 to 8.1±0.8×10^4, 1.1±0.6 to 2.2±1.1×10^5 and 1.0±0.7 to 1.5±1.2×10^5 CFU/m^2, respectively. The *Salmonella* counts recorded varied from sample type and locations but there was no significant difference ($p>0.05$) recorded. The *Salmonella* counts recorded on knives used in cutting fresh pork, especially in the Mile 1 location were higher while the lowest count was recorded in knives used in cutting fresh Pork in the Creek Road market.

The results of the bacterial load of wastewater from the pork meat sample in Table 4 showed that the mean total heterotrophic bacterial load, total *Salmonella*, *Shigella*, coliform and staphylococcal load was 0.99±0.28 to 1.3±0.7×10^7, 6.0±0.6 to 9.3±0.5×10^4, 7.6±0.8 to 14.3±1.6×10^4, 2.7±1.7 to 4.2±3.4×10^5 and 1.4±0.4 to 3.8±2.2×10^5 CFU/mL. There was no significant difference ($p>0.05$) recorded in the *Salmonella* counts of all the wastewater samples. More so, the highest *Salmonella* counts of the wastewater sample were recorded from Creek Road while the lowest count was recorded in the pork wastewater sample from Rumuolumeni

Table 1: Bacterial Load (CFU/g) of Pork Meat Samples from the various Locations

Sample	THB (×10^7)	T SAL (×10^4)	T SH (×10^5)	TCC (×10^5)	TSC (×10^5)
CHINDA	1.05±0.33[a]	4.1±0.48 [a]	1.2±0.17 [a]	2.1±1.4 [a]	1.4±0.7 [a]
CREEKROAD	1.27±0.51 [a]	6.9±0.99 [a]	1.3±0.18 [a]	3.4±2.11 [a]	1.6±0.9 [a]
EGBELU	1.43±0.55 [a]	3.7±0.45 [a]	0.85±0.11 [a]	2.5±1.9 [a]	2.0±0.22 [a]
IWOFE	1.51±0.49 [a]	6.1±0.94 [a]	1.1±0.19 [a]	2.0±1.12 [a]	1.3±0.5 [a]
MILE ONE	1.35±0.25 [a]	7.4±0.89 [a]	1.8±0.27 [a]	2.5±1.35 [a]	1.4±0.4 [a]
RUMUKPAKANI	1.32±0.74 [a]	5.7±1.44 [a]	1.6±0.28 [a]	7.8±5.1 [b]	3.3±0.6 [a]
RUMUOKORO	1.44±0.31 [a]	4.8±0.64 [a]	1.4±0.19 [a]	6.1±3.2 [b]	2.5±1.8 [a]
RUMUOLUMENI	1.18±0.52 [a]	3.7±0.46 [a]	1.7±0.28 [a]	3.7±1.5 [a]	2.2±1.3 [a]
P-Value	0.342	0.932	0.964	<0.001	0.472

*Means with similar superscript ([a, b, c]) down the column showed no significant difference ($p>0.05$).

Keys: THB: Total Heterotrophic Bacteria; TSAL; Total Salmonella, TSH: Total Shigella, TCC; Total Coliform Count, TSC; Total Staphylococcal count

Table 2: Bacterial load of tables used for slaughter of Pork

Sample	THB (×10^7)	T SAL (×10^4)	T SH (×10^4)	TCC (×10^5)	TSC (×10^5)
Chinda	1.0±0.3 [a]	4.7±0.5 [a]	9.3±1.1 [a]	2.6±0.6 [a]	1.3±0.8 [a]
Creek Road (fresh)	1.8±0.4 [c]	6.3±0.7 [a]	8.9±1.0 [a]	3.0±1.1 [a]	1.7±0.8 [a]
Egbelu (roasted)	1.4±0.5 [abc]	6.4±0.7 [a]	7.6±0.8 [a]	2.2±0.5 [a]	1.7±0.4 [a]
Iwofe (roasted);	1.1±0.3 [a]	3.6±0.5 [a]	4.4±0.7 [a]	2.3±1.1 [a]	1.5±0.7 [a]
Mile 3 (fresh)	1.4±0.5 [abc]	5.3±0.6 [a]	8.7±1.1 [a]	2.4±1.1 [a]	2.4±0.6 [a]
Rumukpakani (roasted)	1.2±0.2 [ab]	4.7±0.6 [a]	9.9±1.1 [a]	2.6±1.5[a]	1.5±0.8 [a]
Rumuokoro (fresh)	1.5±0.4 [bc]	8.1±0.9 [a]	10.3±1.2 [a]	4.2±2.2 [a]	1.8±0.7 [a]
Rumuolumeni (fresh)	1.8±0.2 [c]	7.6±0.8 [a]	7.7±0.8 [a]	3.6±1.6 [a]	1.5±0.5 [a]
P-value	0.002	0.953	0.984	0.131	0.255

*Means with similar superscript ([a, b, c]) down the column showed no significant difference ($p>0.05$)

Keys: THB: Total Heterotrophic Bacteria; TSAL; Total Salmonella, TSH: Total Shigella, TCC; Total Coliform Count, TSC; Total Staphylococcal Count

Table 3: Bacterial load of knife used for slaughter of pork

Sample	THB ($\times 10^7$)	T SAL ($\times 10^4$)	T SH ($\times 10^4$)	TCC ($\times 10^5$)	TSC ($\times 10^5$)
CHINDA	1.2±0.5 [a]	1.5±0.2 [a]	4.7±0.6 [a]	1.1±0.6 [a]	1.1±0.8 [a]
CREEKROAD	1.4±0.6 [a]	1.4±0.2 [a]	2.7 ±0.3[a]	1.2±0.5 [a]	1.0±0.7 [a]
EGBELU	0.91±0.4 [a]	4.4±0.5 [a]	5.1±0.6 [a]	1.9±0.6 [a]	1.1±0.5 [a]
IWOFE	1.2±0.5 [a]	4.8±0.7 [a]	4.3±0.5 [a]	1.2±0.5 [a]	1.2±0.5 [a]
MILE ONE	0.99±0.5 [a]	5.4±0.6 [a]	8.1±0.8 [a]	1.4±0.8 [a]	1.3±0.8 [a]
RUMUKPAKANI	1.5±0.4 [a]	3.7±0.4 [a]	6.4±0.8 [a]	2.2±1.1[a]	1.4±0.4 [a]
RUMUOKORO	1.4±0.6 [a]	5.1±0.6 [a]	5.9±0.7 [a]	1.6±0.2 [a]	1.5±1.2 [a]
RUMUOLUMENI	1.0±0.2 [a]	5.3±0.5 [a]	5.9±0.6 [a]	1.8±0.8 [a]	1.5±0.5 [a]
P-value	0.363	0.735	0.929	0.422	0.901

*Means with similar superscript ([a, b, c]) down the column showed no significant difference (p>0.05)
Keys: Rumukpakani (roasted); Rumuokoro (fresh); Rumuolumeni (fresh); Chinda (roasted); Mile 3 (fresh); Egbelu (roasted); Iwofe (roasted); Creek Road (fresh).

Table 4: Bacterial Load of water used for washing Pork after slaughter

Sample	THB ($\times 10^7$)	T SAL ($\times 10^4$)	T SH ($\times 10^4$)	TCC ($\times 10^5$)	TSC ($\times 10^5$)
CR	1.3±0.2 [a]	9.3±0.5 [a]	9.7 ±1.0[a]	3.2±1.7 [a]	1.9±0.7 [ab]
M	1.3±0.7 [a]	8.3±0.9 [a]	8.6±0.9 [a]	2.7±1.7 [a]	1.6±0.6 [ab]
RB	0.99±0.28 [a]	8.5±0.9 [a]	14.3±1.6 [a]	3.8±2.3 [a]	1.4±0.4 [a]
RC	1.0±0.5 [a]	6.0±0.6 [a]	7.6±0.8 [a]	4.2±3.4 [a]	3.8±2.2 [b]
P-value	0.363	0.735	0.929	0.422	0.041

*Means with similar superscript ([a, b, c]) down the column showed no significant difference (p>0.05)
Keys: RB: Rumuokoro (fresh); RC: Rumuolumeni (fresh); M: Mile 1 (fresh); CR: Creek Road (fresh).

The results of the prevalence of *Salmonella* spp across the locations showed that pork meat samples from Rumuolumeni recorded the highest (20.7%) *Salmonella* prevalence while the least (5.7%) prevalence was recorded from pork meat samples in Iwofe locations (Fig. 1).

Results of the prevalence of *Salmonella* spp in the samples showed that the highest (33.3%) prevalence of *Salmonella* sp was recorded in Rumukpakani roasted pork and Rumuolumeni fresh pork samples while the least (15.15%) prevalence was recorded in the roasted pork from Iwofe sample (Fig. 2).

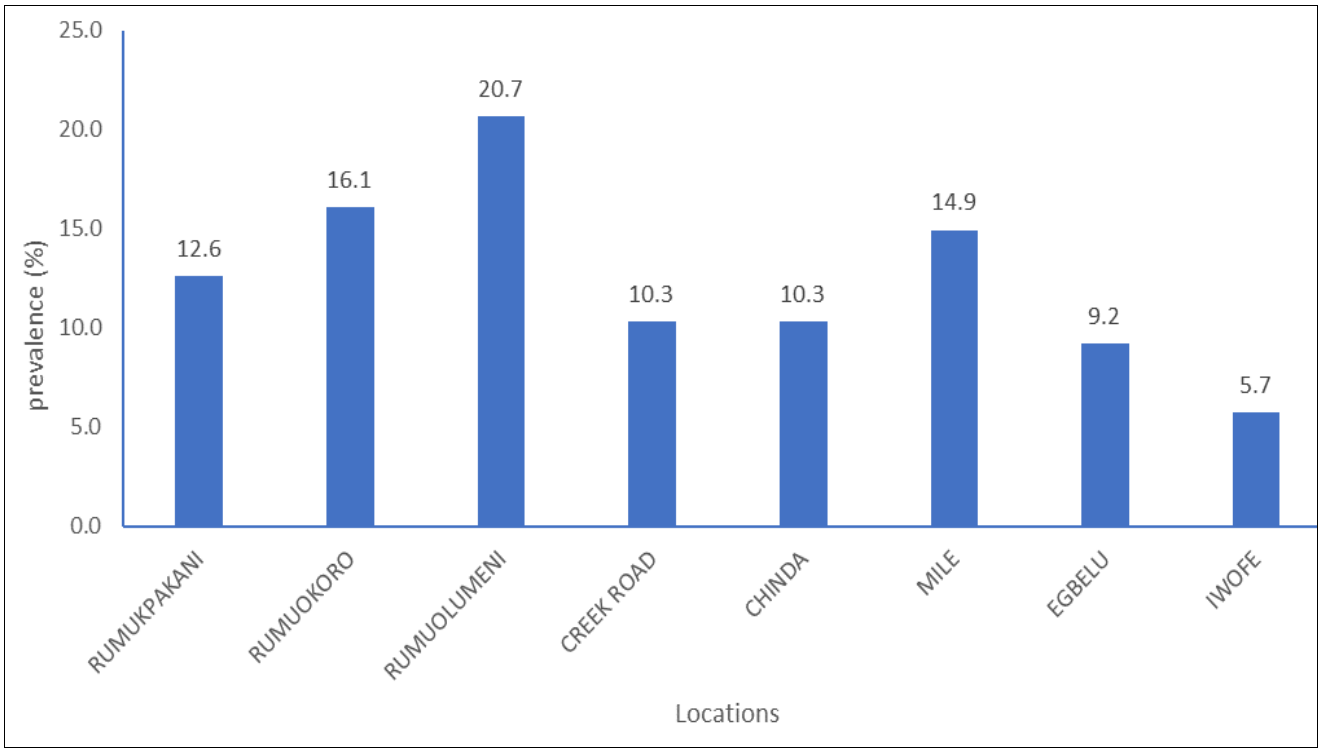

Fig 1: Prevalence of *Salmonella* sp across the locations

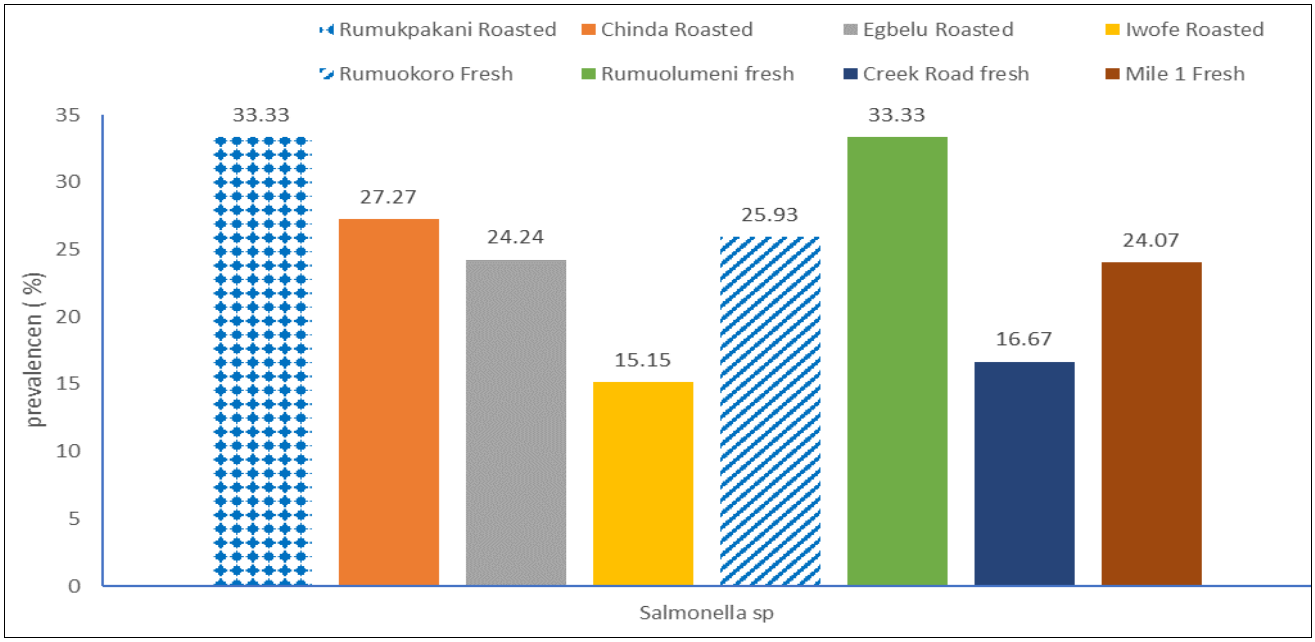

Fig 2: Prevalence of *Salmonella* sp in the samples from the different Locations

Results of the antibiotic susceptibility pattern of *Salmonella* sp isolated from roasted Pork samples showed that isolates were highly resistant to Ampiclox (90.9%), Nalidixic (72.7%), Imipenem/Cilastatin (69.7%), Amoxicillin Clavulanate (66.7%), Cefuroxime (45.5%) and Cefotaxime (45.5%). Ofloxacin (90.9%), levofloxacin (75.8%), Gentamycin (66.7%) and Ceftriaxone /Sulbactam (60.6%) displayed high antibiotic activity against the isolates (Table 5). Results of the antibiotic susceptibility pattern of *Salmonella species* isolated from fresh pork samples were highly resistant to Ampiclox (83.3%), ofloxacin (74.1%), Nalixidic acid and amoxicilline/clavulanate (88.9%) while 100% were susceptible to Levofloxacin (Table 6).

The results of the antibiotic susceptibility pattern of *S. bongori*, *S.* Typhimurium, *S. enterica* and *S. arizonae* is presented in Table 7a while the antibiotic susceptibility pattern of *S. typhi*, *S. diarizonae* and *S. enteritidis* is presented in Table 7b.

The results of the multiple antibiotics-resistant index of the isolates showed that 100% of the isolates had a MAR index greater than 0.2.

(Table 8). Results further showed that all the different species and serovars of *Salmonella* displayed varying degrees of MARi.

Table 5: Susceptibility Pattern of *Salmonella* sp Isolated from Fresh Pork Samples (N=54)

Antibiotics	% Resistant	% Intermediate	% Susceptible
Cefuroxime	30 (55.6)	4 (7.4)	20 (37.0)
Ampiclox	45 (83.3)	3 (5.6)	6 (11.1)
Cefotaxime	24 (44.4)	0	30 (55.6)
Imipenem/Cilastatin	44 (81.5)	0	10 (18.5)
Ofloxacin	40 (74.1)	0	14 (25.9)
Gentamycin	34 (63)	0	20 (37)
Nalidixic	48 (88.9)	2 (3.7)	4 (7.4)
Levofloxacin	0	0	54 (100)
Ceftriaxone Sulbactam	19 (35.2)	5 (9.3)	30 (55.6)
Amoxicilin Clavulanate	48 (88.9)	3 (5.6)	3 (5.6)
Cefixime	13 (24.1)	6 (11.1)	35 (64.8)
Nitrofurantoin	25 (46.3)	6 (11.1)	23 (42.6)

Table 6: Susceptibility Pattern of *Salmonella* sp Isolated from Roasted Pork Samples (N=33)

Antibiotics	% Resistant	% Intermediate	% Susceptible
Cefuroxime	15 (45.5)	10 (30.3)	8 (24.2)
Ampiclox	30 (90.9)	2 (6.1)	1 (3.0)
Cefotaxime	15 (45.5)	2 (6.1)	16 (48.5)
Imipenem/Cilastatin	23 (69.7)	7 (21.2)	3 (9.1)
Ofloxacin	3 (9.1)	0	30 (90.9)
Gentamycin	11 (33.3)	0	22 (66.7)
Nalidixic	24 (72.7)	7 (21.2)	2 (6.1)
Levofloxacin	6 (18.2)	2 (6.1)	25 (75.8)
Ceftriaxone /Sulbactam	12 (36.4)	1 (3.0)	20 (60.6)
Amoxicilin Clavulanate	22 (66.7)	10 (30.3)	1 (3.0)
Cefixime	14 (42.4)	10 (30.3)	9 (27.3)
Nitrofurantoin	14 (42.4)	11 (33.3)	8 (24.2)

Table 7a: Antibiotics susceptibility pattern of different *Salmonella* spp.

Antibiotics (Concr.)	*S. Bongori* (N=11)			*S. Typhimurium* (N=17)			*S. Enterica* (N=24)			*S. Aizonae* (N=8)		
	R	I	S	R	I	S	R	I	S	R	I	S
CXM (30μg)	6 (85.7)	0	1 (14.3)	9 (52.9)	3 (17.6)	5 (29.4)	11 (45.8)	4 (16.7)	9 (37.5)	4 (50)	1 (12.5)	3 (37.5)
ACX (10μg)	7 (100)	0	0	14 (82.4)	1 (5.9)	2 (11.8)	20 (83.3)	2 (8.3)	2 (8.3)	7 (87.5)	0	1 (12.5)
CTX (25μg)	4 (57.1)	0	3 (42.9)	8 (47.1)	1 (5.9)	8 (47.1)	9 (37.5)	0	15 (62.5)	6 (75)	0	2 (25)
IMP (10/10μg)	5 (71.4)	2 (28.6)	0	13 (76.5)	1 (5.9)	3 (17.6)	15 (62.5)	3 (12.5)	6 (25.0)	8 (100)	0	0
OFX (5μg)	1 (14.3)	0	6 (85.7)	11 (64.7)	0	6 (35.3)	17 (70.8)	0	7 (29.2)	7 (87.5)	0	1 (12.5)
GN (10μg)	3 (42.9)	0	4 (57.1)	7 (41.2)	0	10 (58.8)	14 (58.3)	0	10 (41.7)	5 (62.5)	0	3 (37.5)
NA (30μg)	6 (85.7)	0	1 (14.3)	12 (70.6)	3 (17.6)	2 (11.8)	21 (87.5)	1 (4.2)	2 (8.3)	6 (75)	2 (25)	0
LBC (5μg)	1 (14.3)	0	6 (85.7)	1 (5.9)	0	16 (94.1)	2 (8.3)	0	22 (91.7)	1 (12.5)	0	7 (87.5)
CRO (45μg)	2 (28.6)	0	5 (71.4)	9 (52.9)	2 (11.8)	6 (35.3)	9 (37.5)	2 (8.3)	13 (54.2)	1 (12.5)	1 (12.5)	6 (750
AUG (30μg)	6 (85.7)	1 (14.3)	0	12 (70.6)	4 (23.5)	1 (5.9)	16 (66.7)	6 (25)	2 (8.3)	7 (87.5)	0	1 (12.5)
ZEM (5μg)	1 (14.3)	0	6 (85.7)	6 (35.3)	3 (17.6)	8 (47.1)	8 (33.3)	4 (16.7)	12 (50)	3 (37.5)	3 (37.5)	2 (25)
NF (300μg)	6 (85.7)	0	1 (14.3)	8 (47.1)	3 (17.6)	6 (35.3)	8 (33.3)	6 (25)	10 (41.7)	6 (75)	0	2 (25)

Key: Cefuroxime (CXM), Ampiclox (ACX), Cefotaxim (CTX), Imipenem/Cilastatin (IMP), Ofloxacin (OFX), Gentamycin (GN), Nalidixic (NA), Levofloxacin (LBC), Ceftriaxone Sulbactam (CRO), Amoxicilin Clavulanate (AUG), Cefexime (ZEM), Nitrofurantoin (NF).

Table 7b: Antibiotics susceptibility pattern of different *Salmonella* spp

Antibiotics (Concr.)	*S. Typhi* (N=11)			*S. Diarizonae* (N=11)			*S. Enteridis* (N=9)		
	R	I	S	R	I	S	R	I	S
CXM (30μg)	6 (54.5)	1 (9.1)	4 (36.4)	5 (45.5)	2 (18.2)	4 (36.4)	4 (44.4)	1 (11.1)	4 (44.4)
ACX (10μg)	10 (90.9)	1 (9.1)	0	10 (90.9)	0	1 (9.1)	7 (77.8)	1 (11.1)	1 (11.1)
CTX (25μg)	4 (36.4)	0	7 (63.6)	5 (45.5)	0	6 (54.5)	3 (33.3)	1 (11.1)	5 (55.6)
IMP (10/10μg)	9 (81.8)	0	2 (18.2)	9 (81.8)	1 (9.1)	1 (9.1)	8 (88.9)	0	1 (11.1)
OFX (5μg)	4 (36.4)	0	7 (63.6)	8 (72.7)	0	3 (27.3)	5 (55.6)	0	4 (44.4)
GN (10μg)	4 (436.4)	0	7 (63.6)	7 (63.6)	0	4 (36.4)	5 (55.6)	0	4 (44.4)
NA (30μg)	11 (100)	0	0	9 (81.8)	1 (9.1)	1 (9.1)	7 (77.8)	2 (22.2)	0
LBC (5μg)	0	1 (9.1)	10 (90.9)	1 (9.1)	0	10 (90.9)	1 (11.1)	0	8 (88.9)
CRO (45μg)	3 (27.3)	1 (9.1)	7 (63.6)	4 (36.4)	0	7 (63.6)	3 (33.3)	0	6 (66.7)
AUG (30μg)	11 (100)	0	0	9 (81.8)	2 (18.2)	0	9 (100)	0	0
ZEM (5μg)	3 (27.3)	1 (9.1)	7 (63.6)	3 (27.3)	3 (27.3)	5 (45.5)	3 (33.3)	2 (22.2)	4 (44.4)
NF (300μg)	4(36.4)	3(27.3)	4(36.4)	3 (27.3)	5 (45.5)	3 (27.3)	4(44.4)	0	5(55.6)

Key: Cefuroxime (CXM), Ampiclox (ACX), Cefotaxim (CTX), Imipenem/Cilastatin (IMP), Ofloxacin (OFX), Gentamycin (GN), Nalidixic (NA), Levofloxacin (LBC), Ceftriaxone Sulbactam (CRO), Amoxicilin Clavulanate (AUG), Cefexime (ZEM), Nitrofurantoin (NF).

Table 8: MAR Index of *Salmonella* Isolates

Mari	*Salmonella* sp (N=87)	*S. Enteriditis* (N=9)	*S. Bongori* (N=11)	*S. typhi* (N=11)	*S. diarizonae* (N=11)	*S. arizonae* (N=8)	*S. enterica* (N=24)	*S. typhimurium* (N=17)
0.1	0.0	0	0	0	0	0	0	0
0.2	4 (4.6)	0	0	1(9.1)	0	1(12.5)	0	1(5.9)
0.3	9 (10.4)	2(22.22)	1(9.1)	2(18.18)	0	1(12.5)	2(8.33)	1(5.9)
0.4	16 (18.3)	2(22.22)	0	2(18.18)	2(28.57)	0	5(20.83)	5(29.4)
0.5	17 (19.5)	2(22.22)	4(36.36)	1(9.1)	1(14.29)	0	12(50.0)	2(11.8)
0.6	14 (16.00	2(22.22)	3(27.27)	3(27.27)	2(28.57)	1(2.5)	0	3(17.7)
0.7	12 (13.8)	0	1(9.10	1(9.1)	1(14.29)	3(37.5)	4(16.2)	4(23.5)
0.8	13 (14.9)	0	0	1(9.1)	1(14.29)	2(25)	1(4.2)	1(5.9)
0.9	1 (1.2)	0	1(9.1)	0	0	0	0	0
1.0	1 (1.2)	1(11.11)	0	0	0	0	0	0

Discussion

Food safety hazards caused by food-borne pathogens such as *Salmonella* sp remain a major problem for the food industry. Salmonellosis is an important health problem and a major challenge worldwide having greater significance in developing countries (Chaudhary *et al.*, 2015) [8]. The bacterial load in the pork meat (fresh and roasted) and contact surfaces were very high. The total heterotrophic bacterial load and *Salmonella* counts of the fresh pork meats were higher than those obtained in the roasted (ready-to-eat) pork meat samples. In a previous study, the acceptable limit of bacterial contamination of foods as recommended by The International Microbiological Standards for Total Bacterial plate counts was 1.0×10^5 CFU/g (Zakki *et al.*, 2017) [21]. Thus, the bacterial counts obtained in the present study exceeded this limit. Although the counts varied across the samples, findings showed that there were no significant differences ($p>0.05$) in both the total heterotrophic bacterial load and *Salmonella* load. More so, results showed that the fresh pork meat samples bought from the Mile 1 market had the highest *Salmonella* counts compared to those recorded in other locations. The high bacterial load and *Salmonella* load recorded in the present study might not be limited to environmental contaminations but unhygienic practices such as the use of contaminated water for washing meats and contact surfaces during slaughtering as well as contaminated packaging materials. This agreed with Dang-Xuan *et al.*, (2016) [10] who suggested that microbial contamination in pork can occur at any stage, from slaughtering to distribution of pork at retail or pork handling in the households.

Comparable to the present study, Ngo *et al.*, (2021) [17] reported high total bacterial and *Salmonella* counts which exceeded 5.7 $\log_{10}$CFU/g. They also reported variations in the counts based on locations which agreed with the present study. Although the present study showed no significant differences ($p>0.05$) in the THB and *Salmonella* counts obtained in the different locations, one cannot rule out the impact of the environment on the contamination of food products. For instance, slaughterhouses sited in dumpsites could be prone to microbial contaminations compared to those sited in areas not close to dumpsites. In agreement with the present study which showed that fresh pork had a higher bacterial load than roasted pork, Orjiakor *et al.*, (2021) [18] reported that the total bacteria count of smoked (roasted) pork ranged from 2.2×10^4 - 9.0×10^4 while the counts for fresh pork ranged from 1.0×10^6 - 6.0×10^6 cfu/g. The low counts recorded in the roasted pork as compared to the fresh pork could be attributed to the effect of the smoke. In a previous study, it was reported that smoking meat improves desirable flavour, and colour, slows down the onset of oxidative rancidity and microbial invasion-induced spoiling, dries the meat and that the combined antimicrobial and antioxidant properties of formaldehyde, carboxylic acids, and phenols in smoke could be responsible for all of the positive effects of smoking (Bhuyan *et al.*, 2018) [7].

Eighty-seven *Salmonella species* were isolated from 113 pork samples. Out of these, fifty-four (62%) were isolated from fresh pork meat while thirty-three (37.9%) were isolated from roasted pork. Previous studies have reported the presence of *Salmonella* sp in pork samples. Orjiakor *et al.*, (2021) [18] in their study amongst other bacteria isolated *Salmonella* sp in roasted and fresh pork samples. Although the prevalence of *Salmonella* sp in their study was less compared to the present study. The variations in *Salmonella* prevalence observed in the present study could be attributed to the quantity of samples and other handling or processing techniques including geographical locations (Zhou *et al.*, 2019) [22]. The low prevalence of roasted pork over fresh pork agreed with the present study. Ngo *et al.*, (2021) [17] reported a high prevalence of *Salmonella* sp in raw pork sold by street food vendors and traditional markets which agreed with the present study. *Salmonella enterica* non-typhoidal serovars, like serovar Typhimurium (S. Tm), are a major global cause of foodborne illnesses that result in hospitalisations and fatalities (Anderson and Kendall, 2017) [4] while *Salmonella enterica* serovar Typhi, is the cause of typhoid in humans (Dougan and Baker, 2014) [1].

One important factor in the spread of antibiotic resistance is food products (Gugu *et al.*, 2015) [13]. The *Salmonella* isolates displayed multi-drug resistance. The isolates were highly resistant to Ampiclox, Nalidixic acid, Amoxicillin/ Clavulanate, Imipenem and cefuroxime. Although they also exhibited resistance against gentamycin, ofloxacin and nitrofurantoin, they were highly susceptible to levofloxacin with only 6.9% resistance recorded for levofloxacin. The high resistance observed in the present study contradicts the findings of Aslama *et al.*, (2012) [6] who reported that all 29% *Salmonella* isolates from meat were susceptible to amoxicillin-clavulanic, ceftriaxone, ampicillin, gentamicin, and nalidixic. Wang *et al.*, (2021) [19] in their study reported high prevalence of *Salmonella* sp and that the isolates were highly resistant to ampicillin/ampiclox. This high resistance observed in ampiclox agreed with the present study. Due to ampicillin's widespread clinical use, *Salmonella* resistance to the antibiotic has become widespread in both Africa and the USA (Ke *et al.*, 2014) [16]. More so, the present study contradicts Wang *et al.*, (2021) [19] who reported that only one (6.3%) of their *Salmonella* isolates were resistant to ofloxacin a fluroquinolone antibiotics whereas in the present study, 34 isolates with a prevalence of 39.1% were resistant to ofloxacin. Smith *et al.*, (2016) reported that all *Salmonella* isolates in their study were completely susceptibility to ofloxacin; however, in the present study, 60% of the isolates were resistant to ofloxacin. According to Ou *et al.*, (2020), fluoroquinolone- resistant isolates have emerged and are at high levels of resistance.

The high level of antibiotics resistant observed in the present study could be attributed to the unscientific use of broad-spectrum and cheap antimicrobials in the rearing of livestock resulting in an increased number of antibiotic-resistant and multidrug resistant (MDR) isolates (Wang *et al.*, 2021) [19]. It is noteworthy that both the *Salmonella* isolates from the fresh pork and roasted pork displayed MDR with MAR index greater than 0.2. This could be a public health concern especially since this roasted pork are mostly consumed without further preparation. Thus, transfer of resistant isolates to consumers is imminent.

Conclusion

The bacterial load in the pork meats were generally high and the antibiogram showed multi-drug resistance especially on aminoglycosides and fluroquinolones which are known as the last resort for *Salmonella* infections. More so, the high prevalence of *Salmonella* in the pork both fresh and roasted implied poor hygienic processing methods of pork meats which could serve as a vehicle for *Salmonella* transmission to pork consumers.

Thus, proper and stringent processing methods should be implored in the preparation of pork meat before consumption to eliminate bacterial as well as salmonella loads.

Conflict of Interest: Not available

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