Comparative Studies on Bacteriology and Antibiogram of Isolates from Ready-To-Eat and Raw Meat Samples

N. P. Akani, Chinonyerem Chikwendu Jr., S.A. Wemedo

Department of Microbiology, Rivers State University, Nkpolu- Oroworukwo, Port Harcourt, Rivers State, Nigeria

Abstract: **This research looks at the comparative studies on bacteriology and antibiogram of isolates from raw and ready-to-eat (RTE) meat samples in Port Harcourt Nigeria. Ninety (90) samples were collected from three markets in Port Harcourt and subjected to microbiological procedures. The results recorded no significant difference (p>0.05) of Total heterotrophic bacteria count (THBC) in RTE from various locations. THBC ranged between 7.50±1.94 and 5.74±1.35 at Mile 1 market and 6.98±2.15 and 6.04±1.55 at Rumueme market. The *Salmonella-Shigella* count of the ready to eat meat ranged from 3.26±.69 to 3.16±.98 at Mile 3 market and 2.40±.66 to 2.04±.84 at Rumueme market and indicates no significant difference (p>0.05) The coliform count of beef across the location showed significant differences (p≤0.05). While counts of beef in Rumueme market was higher and ranges (3.38±.86 -2.20±.78) than those of the Mile 1 and Mile 3 market. Total Staphylococcal count (TSC) showed no significant difference (p≥0.05) with counts of RTE ranging Mile 1 market (7.50±1.94to 7.22±2.25) Mile 3 (7.68±1.60 to 7.02±2.00) and Rumueme (6.98±2.15 to 6.04±1.55) and a total of 52 bacterial isolates with vary percentage of occurance such as *Staphylococcus* sp 12(23.07%), *Bacillus* sp 9(17.30%), *E. coli* 5*(9.61%), Enterobacter* sp 4(7.69%), *Jeotgalicoccus pinnipedialis 7(13.46%), Macrocuccus caseolyticus 2(3.38%), Klebsiella sp 3(5.76%), Morganella morgani 3(5.76%), Pragia fontium 3(5.76%), Tatumella ptyseos 2(3.84%), Pectobacterium wasabiae 2 (3.84%).* **Consequently, *Staphylococcus* sp, *Bacillus* sp, *E. coli, and Enterobacter* sp showed high resistance to antibiotics such as, Augumentin, vancomycin, ceftazidine, cloxacillin, Erytromycin, cefuroxine, and ceftriaxone while Staphylococcus was susceptible at (66.67%) to gentamycin and ofloxacin (88.33%) and other isolates were 100% susceptible to ofloxacin. Molecular identification of 3 isolate using PCR confirmed *S. aureus* at 65.8% and *Lycinibacillus macroides* at 100%. The megaA, and VanB genes were indentified in 2 Staphylococcus spp while AAIC gene was identified in *Lycinibacillus macrolides.* All three isolates had plasmid at 10kbp. The 52 isolates had 100% multidrug resistance index of more than 0.2. and were 100% multidrug resistant. Public health awareness campaigns are advocated to sensitize meat sellers and consumers in order to mitigate or eliminate several health issues emanating from unhygienic meat slaughtering, preparation and consumption.**

Key Words: **Multiple Antibiotic Resistance (MAR), antimicrobial susceptibility, molecular identification, Raw and Ready to Eat, meat.**

I. INTRODUCTION

Raw meat for the most part alludes to an uncooked muscle tissue of a creature utilized for food [29]. In meat creation industry, the term 'meat' alludes explicitly to mammalian substance, while the words 'poultry' and 'fish' are utilized to separate between the tissue of winged animals and sea-going animals [29]. While Ready-to-eat meat items are meat or poultry items that come in eatable structures and needn't bother with extra planning or cooking [14]. Although they may receive additional preparation (for example, reheating) for a better taste or appearance. This category few instance of RTE products are hot dogs, Suya meats, cold cuts, kelishi (dry meat), and other deli-style meat and poultry , Meat, either raw or ready to eat (RTE) level are getting status in our day by day life. These nourishments being retailed as bundled nourishments are intended to have a long timeframe of realistic usability, require next to no planning work and are ideal for crisis endurance readiness [32. While new meats are regularly intended to have low time span of usability [32]. Meats are consistently powerless to tainting and ensuing development by food borne microbes (*Salmonella, Enteritidis, Staphylococcus aureus, Bacillus cereus, Listeria monocytogenes*) during their planning. There is a tremendous worry of expanding anti-microbial opposition of these microorganisms[18], [27].). Utilization of regular antibacterial compound, for example, concentrates of flavors and spices and so on., for food protection is getting tremendous enthusiasm among scientists [29]. [9].,[24].

Antimicrobial specialists are for the time being the world's just any expectation of disposing of irresistible sicknesses. Notwithstanding, the adjustment in example of obstruction of pathogenic organisms to fundamental anti-infection agents, particularly multidrug safe once has reduced the adequacy of known anti-toxins [25]. As the frequencies of obstruction are expanding around the world, this represents an intense peril to advancement of good wellbeing and a wide range of anti-toxins, including the significant last-dump sedate [5].. Consequently, there is requirement for assessing elective likely restorative specialists with antimicrobial properties. The restorative estimations of plants lie in the synthetic substances presents in the pieces of the plant, for example, seed, leaves bark and root. These substances produce unequivocal physiological activity in the human body. Antimicrobial

opposition in food borne microorganisms is of noteworthy worry to human wellbeing. [4], [27. This is because of the way that a considerable lot of the medications that are utilized to treat human diseases are utilized in creature cultivation as prophylactics and feed supplements, which have been appeared to the determination of safe confines that may influence human wellbeing on the off chance that they get into the natural way of life [32].There has been a connection appeared between the expanded utilization of specific anti-toxins and expanded protection from these anti-infection agents [32], [30]. This study is aimed at evaluating the bacteriology of ready to eat meats in different markets and to develop an antibiogram using standard antibiotics

II. METHODOLOGY

Collection of Ready to Eat Meat

Total of ninety (90) ready to eat meat (beef, chicken and goat) were bought from vendors. Samples were collected weekly for a period of three months from the three different locations (Mile 1 market, Rumueme (Mile 4) Market, and Mile 3 Market) in River State, Nigeria. The samples were collected in well labeled sterile containers and then put into an ice-chest. Samples were immediately transported to the Microbiology laboratory of the Rivers State University for analysis.

Microbiological Analysis of Samples

Stock analytical unit was set up by gauging 10 grams of prepared to eat meat tests separately and homogenizing in 90ml of clean typical saline. Ten times sequential weakening strategy was proceeded by pipetting 1ml of the example into 9ml of clean ordinary saline up to 6 weakenings (weakening element from 10^{-1} to 10^{-6}). After sequential weakening, two weakening elements (10^{-2} and 10^{-4}) were immunized in copies into previously arranged sterile plates of "Mannitol Salt Agar, Salmonella Shigella Agar, Eosin Methylene Blue Agar and 10^{-3} and 10^{-4} of Nutrient Agar plates using spread plate technique. The inoculated plates were incubated at 37℃ for 16 to 24 hours after which growths were counted and analyzed" [19]. Isolates were purified and stored in nutrient agar slants for further analysis. The pure isolates were identified using the tests and methods described in [6]. Such test include; gram staining, motility test, sugar fermentation, catalase, oxidase, methyl red, indole,voges-proskauer, citrate utilization, heamolysis, coagulase, and starch hydrolysis.

TABLE OF IDENTIFICATION OF ISOLATES

		Microscopy			Microscopy									Sugar fermentation									
S/N	Isolate	Surface	Elevation	Colour	Gram rxn	Shape	Catalase	Oxidase	Indole	MR	VP	Citrate	Haemolysis	Motility	Coagulase	Glucose	Lactose	Arabinose	Arafinose	Mannitol	Sucrose	Fructose	Probable organism
1	GTe	rough	Flat	Cream	+ve	Cocci	+	+	-	+	-	+	γ	+	-	-	-	-	-	-	+	+	*Jeotgalicoccus pinnipedialis*
2	GTc	Smooth	Flat	Cream	+ve	Cocci	+	'	-	+	-	-	γ	+	+	+	+	+	+	-	-	+	*Staphylococcus Sp*
3	BFe	Smooth	raise	White	-ve	Rods	+	-	-	-	+	+	α	-	+	+	+	+		+	+		*Klebsiella pneumoniae*
4	BFa	Rough	Flat	Cream	+ve	Cocci	+	+	-	+	-	-	γ	-	+	+	+	-	-	-	+	+	*Staphylococcus hyicus*
5	BFc	Smooth	Flat	Cream	-ve	Rods	+	+	-	+	-	+	γ	+		+	+	-	-	-	-	+	*Pragia fontium*
6	GT-11	Smooth	raise	Purple	-ve	rods	+	-	+	+	-	-	γ	+	+	+	+	-	+	+	+	+	*Morganella morgani*
7	GTer	Rough	raise	White	+ve	Cocci	+	+	-	+	-	+	γ	+		+	+	+	+	+	+	-	*Staphylococcus* sp
8	CH1-	rough	Flat	White	+ve	Cocci	+	+	+	-	+	+	α	+	-	+	+	-	-	-	-	-	*Staphylococcus epidermidis*
9	GTd	Smooth	Flat	Pale yellow	+ve	Rods	+	+	-	-	-	+	α	-	-	-	-	-	-	-	-	-	*Lysinibacillus sphaericus*
10	CHd	rough	Flat	Cream	+ve	Cocci	+	+	-	+	-	+	γ	+	-	-	-	-	-	-	+	+	*Jeotgalicoccus pinnipedialis*
11	BF2-	Smooth	Flat	Cream	-ve	Rods	+	'	-	-	-	-	α	-	-	+	-	-	+	-	+	+	*Tatumella ptyseos*
12	GTb	rough	Flat	White	+ve	Cocci	+	-	+	-	-	+	γ	-	-	+	+	-	-	+	+	+	*Staphylococcus napalensis*
13	GT-12	rough	Flat	Cream	+ve	Cocci	+	+	-	+	-	+	γ	+	-	-	-	-	-	-	+	+	*Jeotgalicoccus pinnipedialis*

14	CH-1a	Smooth	Flat	White	+ve	Rods	+	+	+	+	+	+	β	+	+	+	+	+	+	+	+	+	*Bacillus* tequilensis
15	Cha	Smooth	raise	Yellow	+ve	Rods	+	+	+	-	-	+	α	-	+	+	+	-	-	+	+	+	*Enterobacter asburiae*
16	CHc	Smooth	raise	Purple	-ve	rods	+	-	+	+	-	-	γ	+	+	+	+	+	+	+	+	+	*Escherichia coli*
17	CH-22	Rough	Flat	Cream	+ve	Rods	+	+	-	+	-	+	γ	+	-	+	+		-	+	-	+	Brevibacillus brevis
18	BFd	Rough	Flat	Cream	+ve	Rods	+	+	-	+	-	+	γ	+	-	+	+		-	+	-	+	Brevibacillus brevis
19	BF-1	Smooth	raise	Cream	-ve	Rods	+	+	-	+	-	+	γ	+	-	+	+		-	+	+	+	*Pectobacterium wasabiae*
20	Bfbr	Smooth	raise	Purple	-ve	rods	+	-	+	+	-	-	γ	+	+	+	+	-	+	+	+	+	*Morganella morgani*
21	BF-12	rough	Flat	Cream	-ve	Rods	+	+	-	-	+	+	α	+	-	+	-		+	+	+	+	*Enterobacter pyrinus*
22	GT-11r	Smooth	raise	Cream	-ve	Rods	+	+	-	+	-	-	γ	+	+	+	+		+	-	-	+	*Enterobacter asburiae*
23	CH1r	rough	Flat	White	+ve	Cocci	+	+	+	-	+	+	α	+	-	+	+	-	-	-	-	-	*Staphylococcus epidermidis*
24	GTr	Smooth	Flat	Pale yellow	+ve	Rods	+	+	-	-	-	+	α	-	-	-	-	-	-	-	-	-	*Lysinibacillus sphaericus*
25	CHDr	rough	Flat	Cream	+ve	Cocci	+	+	-	+	-	+	γ	+	-	-	-	-	-	-	+	+	*Jeotgalicoccus pinnipedialis*
26	BF-2r	Smooth	Flat	Cream	-ve	Rods	+	'	-	-	-	-	α	-	-	+	-		-	-	+	+	*Tatumella ptyseos*
27	GTbr	rough	Flat	White	+ve	Cocci	+	-	+	-	-	+	γ	-	-	+	+		-	+	+	+	*Staphylococcus napalensis*
28	GT-12r	rough	Flat	Cream	+ve	Cocci	+	+	-	+	-	+	γ	+	-	-	-	-	-	-	+	+	*Jeotgalicoccus pinnipedialis*
29	CHA3r	Smooth	Flat	White	+ve	Rods	+	+	+	+	+	+	β	+	-	+	+	+	+	+	+	+	*Bacillus* tequilensis
30	Char	Smooth	raise	Yellow	+ve	Rods	+	+	+	-	-	+	α	-	+	+	+	-	-	+	+	+	*Enterobacter asburiae*
31	GTEa	rough	Flat	Cream	+ve	Cocci	+	+	-	+	-	+	γ	+	-	-	-	-	-	-	+	+	*Jeotgalicoccus pinnipedialis*
32	GTc	Smooth	Flat	Cream	+ve	Cocci	+	'	-	+	-	-	γ	+	+	+	+	+	+	-	-	+	*Staphylococcus Sp*
33	Gtc 22	Smooth	raise	White	-ve	Rods	+	-	-	-	+	+	α	-	+	+	+		+	+	+		*Klebsiella pneumonia*
34	BFar	Rough	Flat	Cream	+ve	Cocci	+	+	-	+	-	-	γ	-	-	+	+	-	-	-	+	+	*Staphylococcus hyicus*
35	BFcr	Smooth	Flat	Cream	-ve	Rods	+	+	-	+	-	+		+	-	+	+	-	-	-	-	+	*Pragia fontium*
36	GTer	rough	Flat	Cream	+ve	Cocci	+	+	-	+	-	+	γ	+	+	-	-	-	-	-	+	+	*Jeotgalicoccus pinnipedialis*
37	GTcr	Smooth	Flat	Cream	+ve	Cocci	+	'	-	+	-	-	γ	+	+	+	+	+	+	-	-	+	*Staphylococcus Sp*
38	BFer	Smooth	raise	White	-ve	Rods	+	-	-	-	+	+	α	-	+	+	+		+	+	+		*Klebsiella pneumonia*
39	BFa3r	Rough	Flat	Cream	+ve	Cocci	+	+	-	+	-	-	γ	-	-	+	+	-	-	-	+	+	*Staphylococcus hyicus*
40	BFc3r	Smooth	Flat	Cream	-ve	Rods	+	+	-	+	-	+	α	+	-	+	+	-	-	-	-	+	*Pragia fontium*
41	CH-1Ar	Smooth	raise	Light cream	+ve	Cocci	+	-	+	-	-	+	α	+	+	+	+	+	+	+	+	+	*Macrococcus caseolyticus*
42	Bf -22	Smooth	raise	Light cream	+ve	Cocci	+	-	+	-	-	+	α	+	+	+	+	+	+	+	+	+	*Macrococcus caseolyticus*
43	BFer	Smooth	raise	Purple	-ve	rods	+	-	+	+	-	-	γ	+	+	+	+		+	+	+	+	*Escherichia coli*
44	CHcr	Smooth	raise	Purple	-ve	rods	+	-	+	+	-	-	γ	+	+	+	+		+	+	+	+	*Escherichia coli*

45	GTar	rough	Flat	Cream	+ve	Cocci	+	-	-	-	-	+	α	-	-	+	-		-	-	-	+	*Staphylococcus cohnii*
46	GTdr	rough	Flat	Cream	+ve	Rods	+	+	-	+	-	+	α	+	-	+	-	-	-	-	+	+	*Bacillus* sp
47	GTfr	Smooth	raise	Purple	-ve	rods	+	-	+	+	-	-	γ	+	+	+	+		+	+	+	+	*Escherichia coli*
48	CH-22r	rough	Flat	Cream	+ve	Rods	+	+	-	+	-	+	α	+	+	+	-	-	-	-	+	+	*Bacillus* sp
49	CHdr	Smooth	raise	Purple	-ve	rods	+	-	+	+	-	-	γ	+	+	+	+	+	+	+	+	+	*Escherichia coli*
50	BFdr	rough	Flat	Cream	+ve	Rods	+	+	-	+	-	+	α	-	+	+	-	+	-	-	+	+	*Bacillus* sp
51	Bfc	Smooth	raise	Cream	+ve	Cocci	+	-	-	+	-	+	γ	-	+	+	+	+	+	+	+	+	*Staphylococcus sp*
52	Cha-1	Smooth	raise	Cream	-ve	Rods	+	+	-	+	-	+	γ	+	+	+	+	+	+	+	+	+	*Pectobacterium wasabiae*

Antibiotic Sensitivity Testing

The antimicrobial powerlessness profiles of the confines to ordinary anti-microbials were dictated by the Kirby-Bauer circle dispersion technique (Performance Standards for Antimicrobial Susceptibility Testing; Twenty-First Informational Supplement, 2011) on sterile Mueller-Hinton agar. The surface strong media plate was immunized with bacterial suspension (normalized to the 0.5 McFarland) by cleaning over the agar plate surface; being certain that no zone of the surface is sans left of inoculum. This methodology was rehashed a few times, pivoting the agar plate 60° each an ideal opportunity to guarantee even conveyance of the inoculum to the edge of the agar. The plates were left to dry for 3–5 min to permit retention of any dampness preceding applying the anti-toxin circles with the following concentration Ceftriaxone 30µg, Gentamycin 10 µg, Erytromycin 5µg, Ceftaroxine 30 µg, Cloxacillin 5µg, Ofloxacin 5µg, Augmentin 30 µg, Ceftazidine 30 µg, Vancomycin 30 µg. The antibiotic disks were aseptically positioned on the outside of the immunized agar plate with sterile forceps. Each circle was pushed down to guarantee full contact with the outside of the agar. In any event 24 mm was left between the focuses of the circles, and at least 15 mm from the fringe of the plate as well. The plates were then reversed and set in a hatchery inside 15 min of applying the circles. At last, the plates were brooded for 24 h in the incubator at 35°C [7].

Determination of Multiple Antibiotic Resistance Index

Multiple antibiotic resistance indices in relation to this study is referred to as the opposition of bacterial species disconnect to at least three anti-microbials (Davis *et al.,* 2016). Multiple antibiotic resistance (MAR) index was learned for each segregate by utilizing the equation:

MAR = a/b, where,

a = "The number of antibiotics to which the isolate depicted resistance and

b = The total number of antibiotics to which the test isolates has been evaluated for susceptibility" [28]., [23].

Statistical Analysis

Descriptive statistics was utilized to sum up all data got. Analysis Of Variance (ANOVA) was done to test for critical distinction in the total heterotrophic bacteria count, total Coliform count, total *E. coli* count, *Salmonella-shigella* count, total *Staphylococcus* count in the various markets and between the fresh and ready to eat meats. Where there was significant difference, Duncan Multiple Range Test (DMRT) was used to separate the means.

III. RESULTS AND DISCUSSION

The results from Table 1 showed the distribution of total heterotrophic bacteria count of microbial population in various markets for raw and ready to eat meats samples and indicate no significant difference generally. The *Salmonella-Shigella* count of the raw and ready to eat meat is presented in Table 2. Despite the presence of *Salmonella-Shigella* load in the meat samples across the different locations, there was no significant differences in the count except for raw chicken samples in mile 1 and mile 3 markets.

Table 1: variation in total heterotrophic bacterial (thb) count of raw and rte meat sample from various markets sampled

RAW $x10^5$ $cufg^{-1}$				READY TO EAT(RTE) $x10^5$ $cufg^{-1}$		
Markets	Beef	Chicken	Goat	Beef	Chicken	Goat
Mile 1	8.70 ± 1.48^{a}	13.12 ± 1.91^{a}	13.02 ± 2.90^{a}	7.50 ± 1.94^{a}	5.74 ± 1.35^{a}	7.22 ± 2.25^{a}
Mile3	14.20 ± 4.77^{b}	12.88 ± 3.22^{a}	12.14 ± 1.72^{a}	7.02 ± 2.00^{a}	7.56 ± 1.51^{a}	7.68 ± 1.60^{a}
Rumueme	8.42 ± 1.73^{a}	11.08 ± 1.97^{a}	10.72 ± 1.74^{a}	6.98 ± 2.15^{a}	6.54 ± 2.66^{a}	6.04 ± 1.55^{a}

KEY: Means with the same superscript alphabets across the column indicate no significant difference ($p>0.05$)

Table 2: variation in total salmonella/shigella count of raw and ready to eat meat sample from various markets sampled

RAW $x10^5$ $cfug^{-1}$				READY TO EAT(RTE) $x10^5$ $cfug^{-1}$		
Markets	Beef	Chicken	Goat	Beef	Chicken	Goat
Mile 1	5.54 ± 2.05^{a}	7.14 ± 1.52^{b}	5.44 ± 1.95^{a}	$2.92\pm.82^{a}$	$2.20\pm.60^{a}$	2.70 ± 1.51^{a}
Mile3	5.98 ± 3.01^{a}	7.14 ± 2.65^{b}	6.76 ± 1.57^{a}	$3.16\pm.98^{a}$	2.48 ± 1.00^{a}	$3.26\pm.69^{a}$
Rumueme	3.20 ± 1.37^{a}	$3.20\pm.84^{a}$	4.52 ± 1.13^{a}	$2.40\pm.66^{a}$	$2.04\pm.84^{a}$	$2.24\pm.37^{a}$

KEY: Means with different superscript alphabets across the column indicate significant difference ($p\geq0.05$)

Table 3 represents the results of microbial population in various markets for raw and ready to eat meat samples. Generally, there was a significant difference ($p \leq 0.05$). The coliform count of the beef across the location showed significant differences as well. The coliform count of the ready to eat beef in Rumueme market was higher than those of the Mile 1 and Mile 3, and was significantly different from the coliform counts obtained in ready to eat beef from Mile 1.

Table 3: Variation in Total Coliform Count of Raw and Ready To Eat Meat Sample from Various Markets Sampled

Raw $x10^5 cfug^{-1}$				READY TO EAT(RTE) $x10^5 cfug^{-1}$		
Markets	Beef	Chicken	Goat	Beef	Chicken	Goat
Mile 1	4.72±1.41 [b]	3.60±.51 [a]	4.78±1.03 [a]	1.50±.39 [a]	2.20±.54 [a]	2.70±1.26 [a]
Mile3	4.80±.96 [b]	6.44±1.20 [b]	6.06±1.41 [a]	2.70±1.41 [ab]	1.94±.67 [a]	3.82±3.55 [a]
Rumueme	3.00±.78 [a]	5.62±2.10 [b]	5.20±2.97 [a]	3.38±.86 [b]	2.88±1.08 [a]	2.20±.78 [a]

KEY: Means with different superscript alphabets across the column indicate significant difference ($p \leq 0.05$)

Table 4 represents the results of staphylococcal population in various markets for ready to eat meat samples. There was no observed difference ($p \geq 0.05$). The table showed that ready to eat goat meat had the highest mean of staphylococcal load ($3.12 \pm 1.13 x 10^4$ $cfug^{-1}$) and chicken had the last mean of staphylococcal count ($1.54 \pm 0.67 x 10^4$ $cfug^{-1}$). Consequently, mile 3 market showed the highest mean of staphylococcal load and counts ranged from $1.64 \pm .43 x 10^4$ $cfug^{-1}$ to $2.36 \pm 1.11 x 10^4$ $cfug^{-1}$, respectively among other markets.

The result of the antibiotic susceptibility pattern of gram positive and negative bacterial isolates is presented in Table 5. In Table 6 and 7, the MAR indices of the gram positive and negative isolates are illustrated.

Table 4: variation in total staphylococcal count (tsc) of raw and ready to eat meat sample from various markets sampled

Raw x105 cfug-1				READY TO EAT(RTE) x105 cfug-1		
Markets	Beef	Chicken	Goat	Beef	Chicken	Goat
Mile 1	4.34±1.89 a	4.18±1.38a	3.90±1.24 a	1.88±.13 a	1.54±.67 a	2.56±1.19 a
Mile 3	3.76±1.28 a	3.84±.38 a	4.20±.81 a	2.36±1.11 a	1.64±.43 a	1.94±.74 a
Rumueme	4.10±3.24 a	5.58±2.30a	5.32±2.18 a	1.94±.48 a	1.82±.55 a	3.12±1.13 a

Key: Means with same superscript alphabets across the column shows no difference ($p \geq 0.05$)

Table 5: Susceptibility pattern of tested Gram-negative isolates from raw and ready to eat meat samples in various markets

Antibiotics	Conc.	*E.coli*			*Klebsiella sp*			Enterobacter sp			*Pectobacterium sp*			*Tatumella ptyseos*			*Morganella morgani*			*Pragia fontium*		
		R	I	S	R	I	S	R	I	S	R	I	S	R	I	S	R	I	S	R	I	S
CRX	**30µg**	3(60.00)	1(20)	1(20)	3(100)	0.00	0.00	0.00	0.00	4(100)	1(50)	0.00	1(50)	0.00	0.00	2(100)	1(50)	0.00	1(50)	0.00	0.00	3(100.00)
GEN	**10 µg**	4(80.00)	0.00	1(20)	1(33.3)	1(33.3)	1(33.3)	0.00	0.00	4(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	1(50)	0.00	1(50)	1(33.33)	1(33.33)	1(33.33)
ERY	**5µg**	5(100)	0.00	0.00	2(66.7)	0.00	1(33.3)	3(75)	0.00	1(25)	2(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	3(100)	0.00	0.00
CTR	**30 µg**	5(100)	0.00	0.00	3(100)	0.00	0.00	4(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	2(75)	0.00	1(25.)
CXC	**5µg**	5(100)	0.00	0.00	3(100)	0.00	0.00	4(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	3(100)	0.00	0.00
OFL	**5µg**	2(40.00)	1(20)	2(40)	0.00	0.00	3(100)	0.00	0.00	4(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	3(100)
AUG	**30 µg**	5(100)	0.00	0.00	3(100)	0.00	0.00	4(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	3(100)	0.00	0.00
CAZ	**30 µg**	5(100)	0.00	0.00	3(100)	0.00	0.00	4(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	3(100)	0.00	0.00
VAN	**30 µg**	5(100)	0.00	0.00	3(100)	0.00	0.00	4(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	2(100)	0.00	0.00	3(100)	0.00	0.00

KEY : CRX: Cefuroxime, GEN: Gentamicin, ERY: Erytromycin, CTR: Ceftriaxone, CXC: Cloxacillin, OFL: Ofloxacin, AUG: Augmentin, CAZ: Ceftazidime, and VAN: Vancomycin

Keys: N= number of isolate

Table 6: Susceptibility pattern of tested Gram positive isolates from raw and ready to eat meat samples in various markets

Antibiotics	Conc.	*Staphylococcus sp*			*Bacillus sp*			*Jeotgalicoccus pinnipedialis*			*Macrococcus caseolyticus*		
		R	**I**	**S**	**R**	**I**	**S**	**R**	**I**	**S**	**R**	**I**	**S**
Ceftriaxone	**30µg**	3(25.00)	5(41.67)	4(33.33)	5(55.56)	0.00	4(44.44)	6(75.00)	0.00	2(25.00)	0.00	1(50.00)	1(50.00)
Gentamycin	**10 µg**	2(16.67)	2(16.67)	8(66.67)	0.00	1(11.11)	8(88.89)	2(25.00)	3(37.50)	3(37.50)	0.00	0.00	0.00
Erytromycin	**5µg**	11(91.67)	1(8.33)	0.00	6(66.67)	0.00	3(33.33)	5(62.50)	0.00	3(37.50)	2(100.00)	0.00	0.00
Ceftaroxine	**30 µg**	10(83.33)	2(16.67)	0.00	7(77.78)	2(22.22)	0.00	6(75.00)	0.00	2(25.00)	2(100.00)	0.00	0.00
Cloxacillin	**5µg**	12(100.00)	0.00	0.00	9(100.00)	0.00	0.00	8(100.00)	0.00	0.00	2(100.00)	0.00	0.00
Ofloxacin	**5µg**	2(16.67)	0.00	10(83.33)	1(11.11)	0.00	8(88.89)	1(12.50)	0.00	7(87.50)	0.00	0.00	2(100.00)
Augmentin	**30 µg**	12(100.00)	0.00	0.00	9(100.00)	0.00	0.00	8(100.00)	0.00	0.00	2(100.00)	0.00	0.00
Ceftazidine	**30 µg**	12(100.00)	0.00	0.00	9(100.00)	0.00	0.00	8(100.00)	0.00	0.00	2(100.00)	0.00	0.00
Vancomycin	**30 µg**	12(100.00)	0.00	0.00	9(100.00)	0.00	0.00	8(100.00)	0.00	0.00	2(100.00)	0.00	0.00

KEY : CRX: Cefuroxime, GEN: Gentamicin, ERY: Erytromycin, CTR: Ceftriaxone, CXC: Cloxacillin, OFL: Ofloxacin, AUG: Augmentin, CAZ: Ceftazidime, and VAN: Vancomycin *Keys: N= number of isolates*

Table 7. Multiple antibiotic resistance index of bacterial isolate for gram positive organisms from rte meat samples in various markets

MAR n=31	*Bacillus sp*	*Staphylococcus sp*	*Jeotgalicoccus pinnipedialis*	*Macrococcus caseolyticus*
0.4	2(22.22)	0.00	0.00	0.00
0.5	0.00	2(16.67)	0.00	0.00
0.6	0.00	6(50.00)	2(25.00)	0.00
0.7	3(33.33)	0.00	3(37.50)	2(100.00)
0.8	3(33.33)	2(16.67)	0.00	0.00
0.9	1(11.11)	2(16.67)	3(37.50)	0.00

Note: MAR index values greater than 0.2 indicate high risk source of contamination where antibiotics are often used

Table 8. Multiple antibiotic resistance index of bacterial isolates for gram negative organisms from ready to eat meat sample in various markets

MAR (n=21)	*Klebsiella pneumonia*	*E. coli*	*Enterobacter* sp	*Tatumella ptyseos*	*Pectobacterium wasabiae*	*Morganella morgana*	*Pragia fontium*
0.5	0.00	4(80.00)	0.00	0.00	0.00	0.00	0.00
0.6	0.00	0.00	1(25.00)	0.00	0.00	0.00	0.00
0.7	0.00	1(20.00)	3(75.00)	2(100.00)	1(50.00)	0.00	2(66.67)
0.8	2(66.67)	0.00	0.00	0.00	1(50.00)	2(100.00)	1(33.33)
0.9	1(33.33)	0.00	0.00	0.00	0.00	0.00	0.00

Note: MAR index values greater than 0.2 indicate high risk source of contamination where antibiotics are often us.

The aerobic bacterial load of the ready to eat meats showed that the beef was more disposed to contamination as their mode of preparation is questionable with regards to hygiene level of preparation equipment. This result conforms with [2]. More so, samples from Mile 3 market showed relatively high counts compared to other markets for RTE meats. This may be due to the poor hygiene of slaughter houses, meat handlers, water and utensils used during meat preparation, transportation of meats, hygiene of storage facilities or open display of the meat for sell and the dense population of consumers who touch and talk while trying to purchase meats from this market. The coliform count detected in the ready to eat meat could also be attributed to faecal contamination. *E. coli* is an organism that is part of the normal microflora of the intestinal tract of humans and warm-blooded animals and is a commonly used faecal indicator organism [3]. Its presence indicates direct or indirect contamination of faeces [3]. Thus, "their presence in ready-to-eat foods could be an indication of poor hygiene and sanitation or inadequate heat treatment", this result correlates with [3]. The presence of bacteria in ready to eat meats can result from improperly cooked or fried meats and post processing contamination which can occur especially during handling, sales and transportation of RTE meats to the point or location of sales [22]. According to Microbiological guidelines for Ready to eat Food (2014), the guideline on the interpretation of results of hygiene indicator organisms in ready to eat food state that the satisfactory level for *E. coli* is <20, while the border line is 10^2 -< 10^4 cfu/g and unsatisfactory is >10^4. Hence when compared with the standard guidelines, The counts in this current study showed high contamination of *E. coli* in the ready to eat meat. More so, bacteria such as *Salmonella a*nd *Shigella* have been found to be related with various diseases of man such as gastroenteritis. These finding agrees with FAO/WHO [10]. and states that in developing countries such as Nigeria cholera is prevalent due to the feeding habits of people. "*Staphylococcus aureus* can be routinely isolated from humans and associated environments. As such, the presence of coagulase positive staphylococci (a subgroup of *S. aureus*), is an indication of human contact. Some CPS strains generate a toxin which may cause food poisoning. Though, negligible handling of foods can cause staphylococci being present in foods at low levels" [32].. This is probably not going to be a sanitation concern gave the food is either devoured quickly or

put away under high temperature. Broad taking care of as well as temperature misuse may bring about expanded levels and expanded sanitation hazard if poison creation happens.

Fifty-two (52) bacterial isolates of different genera were recovered from the different sample with various percentage of occurance such as *Staphylococcus* sp 12(23.07%), *Bacillus* sp 10(19.23%), *E. coli* 5*(9.61%) ,Enterobacter* sp 4(7.69%), *Jeotgalicoccus pinnipedialis 7(13.46%), Macrocuccus caseolyticus 1(1.92%), Klebsiella sp 3(5.76%), Morganella morgani 3(5.76%), Pragia fontium 3(5.76%), Tatumella ptyseos 2(3.84%), Pectobacterium wasabiae 2 (3.84%).*, all from ready to eat meat samples in the various markets. The predominance in gram positives are seen in *S. aureus* with (38.71%). In this present study, *S.aureus* was recorded as the most frequently isolated bacteria. This is in conformity with the results of Egbebi *et al.* (2011), Nwakanma *et al*. (2015) and Akani *et al* (2020) who also recorded highest percentage of *S. aureus 23.07%*, followed by *Bacillus* sp 19.23%, *Jeotgalicoccus pinnipedialis* (13.46%) and *Macrococcus caseolyticus* (1.92%). However, prevalence of gram negatives showed that the highest was *E. coli* 5(23.80%) this is in conformity with Elnawawi *et al*. (2012); *Enterobacter* sp 4(7.69*%), Klebsiella pneumonia* 3(5.76%), percentages is related with Gill (2005) and Gibbons et al (2006), while Pragia *fontium* 3(14.29%), *Pectobacterium wasabiae* 2(3.84%), and *Tatumella ptyseos* 2(3.84%) showed relatively less prevalence in the meat samples.

The results of anti-toxin affectability test as deciphered utilizing the Clinical Laboratory Standard Institute Guideline (2015) indicated that all the bacterial types had changing powerlessness to the anti-microbials tried. Results showed that ofloxacin amongst other antibiotics was the most effective on bacteria types (gram positive and negative tried creation it the anti-infection with the most noteworthy viability on the disconnects. It was effective on all 52(100%) of the isolates tested; while Gentamycin was effective to 38 (73.08%) of all isolates and Ceftriaxone 26(50.00%). The two must resistant organism *was Staphylococcus sp* and *Bacillus sp*. *Staphylococcal* resistance may be due to some resistance mechanisms such as enzymatic inactivation of the anti-toxin (penicillinase and aminoglycoside-change catalysts), adjustment of the objective with diminished liking for the anti-microbial (prominent models being penicillin-restricting protein 2a of methicillin-safe S. aureus and D-Ala-D-Lac of peptidoglycan forerunners of vancomycin-safe strains), catching of the anti-toxin (for vancomycin and conceivably daptomycin) and efflux siphons (fluoroquinolones and antibiotic medication). [15]. and [17].This result conforms with[26]. [1]. While resistance arising from *bacillus sp* may be due to the ribosome which is one of the significant focuses in the phone for anti-toxins, including numerous clinically significant anti-infection classes, for instance, the streptogramins, lincosamides, pleuromutilins, and macrolides other reasons may be due to over production of the dipeptide antibiotic bacilysin [31], [16].

According to the Nigerian Federal service of Agriculture, Environment and Health (2017), microscopic organisms opposition in creatures and the earth precise audit of Nigerian writing uncovered that safe microbes are generally recouped from domesticated animals including steers, sheep, goats, camels, pigs and poultry. Likewise, correspondingly elevated levels of safe life forms were seen from nourishments, for example, meats, dairy and vegetables. While it is possible that safe living beings in household creatures could have been procured from human and different sources, the significant levels of antimicrobial deposits in Nigerian meats and the low recuperation of safe life forms from natural life highlight antimicrobial use in rural and veterinary practices as the central driver of obstruction. Safe microbes have likewise been recuperated from assumed consumable, common and waste water destinations. They have been found in soils, aquaculture destinations just as somewhere else in the earth.

Consequences of work in South Africa by [18].) on the antimicrobial opposition of microorganisms from poultry varied from those of this flow research for the three anti-toxins normal to the two investigations. In their examination, [18].. detailed that 87 and 92% of every oxygen consuming specie from retail and butchered chickens, separately, were impervious to penicillin (as dictated by utilizing oxacillin); the greater part of the secludes in this current investigation were impervious to beta lactam. Protection from different anti-infection agents among gram-negative microorganisms followed an ordinary dispersion with a top at protection from seven anti-infection agents. Protection from five or six anti-toxins was distinguished distinctly for gram-negative microbes. The numerous obstruction top for gram-positive microbes was at protection from four anti-infection agents. Nonetheless, four of the gram-positive secludes were impervious to seven anti-infection agents. [18]. revealed comparative information for various anti-infection safe microbes disconnected from retail chicken. The discoveries in this current examination carries the level of secludes with MAR record more prominent or equivalent to 0.2 to 100%. Blemish file esteems more noteworthy than 0.2 demonstrate high hazard wellspring of sullying where anti-infection agents are regularly utilized [23], [8]. The nearness of anti-infection safe living beings on domesticated animals items may have serious ramifications for general wellbeing if meat isn't cooked and taken care of appropriately before utilization. Especially, the exchange of opposition from microscopic organisms in poultry, meat or goat (lamb) to those in people may prompt human sickness brought about by strains that are impervious to generally utilized antibiotics100% of the *bacteria* species isolated in this study are likely to show multiple resistances to antibiotics where the antibiotics used for this study are often used.

IV. CONCLUSION AND RECOMMENDATION

Conclusively this study has demonstrated that several *Enterobacteriaceae*, *Staphylococcus* sp and *Bacillus* sp are common in retailed meat (ready to eat) with increasing resistance which posses great medical threat to meat sellers and consumers. Insufficient awareness about food-borne zoonoses or infections could endanger both retailers and consumer health. Education of the meat retailer's community in Port Harcourt markets in terms of hygienic and sanitary precautions would be a pivotal step towards safer food. The abuse of antibiotics must be checked as this is fast leading the antibiotics to completely lose their efficacy against microorganisms especially *Staphylococcus aureus*, *Bacillus sp* and *Escherichia coli*.

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