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Identity and Antibiogram of Bacterial Isolates from Owerri Modern Abattoir, Imo State, Nigeria

Joy Nkeiruka Dike-Ndudim¹, Roseline Nwanneka Ugwuegbu², Henry Chidozie Amah¹, Chizaram Winners Ndubueze^{1*} and Emeka Simon Anikwo³

¹Department of Medical Laboratory Science, Faculty of Health Sciences, Imo State University, Owerri Nigeria.

²Fabian and Rose Clinic and Diagnostic Centre, MCC/Uratta Road, Owerri, Imo State, Nigeria. ³Department of Human Physiology, Faculty of Basic Medical Sciences, Imo State University, Owerri Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

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Original Research Article

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ABSTRACT

Aim: The identity and antibiogram of bacterial isolates from Owerri modern abattoir in Imo State, Nigeria, was investigated with the aim of determining the bacterial profile of the abattoir and their susceptibility pattern to commonly used antibiotics.

Study Design: Cross sectional study

Place and Duration of Study: This study was carried out in Owerri Modern Abattoir located within Owerri metropolis from June to November, 2020.

Methodology: Questionnaires were used to obtain participants' consent, demographic data and sanitary practices in the abattoir. Samples were taken and bacteriological analysis of the samples done using pour plate method. Disc diffusion antibiotic susceptibility testing and minimum inhibitory concentration were performed after colony counting, identification and characterization of the isolates using standard microbiological and biochemical techniques.

*Corresponding author: E-mail: chizaramwinnersndubueze@gmail.com;

Results: Mean viable bacterial counts were generally high with highest counts from contaminated soil (6.13x10⁶CFU/ml) and least from workers hands (1.17x10⁶CFU/ml). *Escherichia coli* had the highest prevalence (18.0%), with the highest counts from soil (3.10%). *Vibrio cholerae* recorded the least prevalence (0.62%), and was isolated only from washing water. High resistance to antibiotics was observed.

Conclusion: Government authority and the general public are advised to ensure adequate environmental sanitation and proper cooking of meat before consumption in order to mitigate the incidence of infection and antibiotic resistance.

Keywords: Antibiogram; bacteria; abattoir; meat; contamination; antibiotic resistance; waste.

1. INTRODUCTION

Abattoir, otherwise known as slaughter house is a facility where animals are killed for consumption as food products. In Nigeria, abattoir business is an important component of the livestock industry, providing domestic meat supply to over 150 million people and employment opportunities for the teaming population. However, just as it is the case in other developing countries, abattoir industries in Nigeria are not developed [1].

Despite the obvious benefit of abattoir operation to man, which includes provision of meat and other useful by-products, it can be very hazardous to public health in respect to the waste it generates and as source of food poisoning. Contamination of meat in abattoirs and meat stalls often result from contaminated water, unhygienic practices like poor handling, use of contaminated tables to display meat for sale and the use of contaminated knives in cutting operations. The contaminating organisms, however, are derived mainly from the hide of the animals and comprise organisms that originate from stomachs and intestines, which are excreted in their feces [2].

Abattoirs are major sources of pollutants in cities and sub urban areas, when the slaughter wastes are not properly managed and especially discharged into water ways, as such practices can introduce enteric pathogens and excess nutrients into surface water [3]. Critical environmental and public health issues are associated with large scale slaughtering of animals. Inadequate portable water supply and poor conditions of public infrastructures, as well as unhygienic conditions of these abattoirs raise serious public health concern, since hygiene problems are not only limited to slaughtering but also associated with incorrect processing of the animals [4].

Contaminated meat that is not properly cooked is one of the main causes of foodborne illness. The risk of zoonotic infections is also associated with contaminated meat through cross infection [5,6].

Contamination of water bodies from abattoir waste constitutes significant environmental and public health hazards [7,8,1,9]. Bacteria from abattoir waste discharged into water columns can subsequently be absorbed to sediments and when the bottom is disturbed, the sediment releases the bacteria back into the water columns presenting long-term health hazards [10].

In Nigeria, numerous abattoirs dispose their effluents directly into the streams and water ways without any type of treatment and the butchered meat is washed by the same water [11].

2. MATERIALS AND METHODS

2.1 Study Area

This study was carried out in Owerri Modern Abattoir which is located in the South Eastern part of Nigeria.

2.2 Sample Collection and Processing

Questionnaires were used to get information concerning demographic data and sanitary practices in the abattoir. A total of 161 samples were obtained from Slaughter slabs (35), Butchering tables (17), Butchering knives (16), Workers hands (9), Wahing water (22), Rinsing water (9), Contaminated Soil (27), Control soil (12) and Meat (14). The effluent was agitated to obtain a homogeneous solution and 1ml transferred into 9mls of sterile broth to make a tenfold dilution [12]. The swab sticks were dipped in sterile normal saline, before samples were taken. It was then added into 1ml of sterile peptone water before dilution. Twenty-five grams (25g) of representative stock meat sample was weighed aseptically and homogenized in 225ml of 0.1% peptone water using a blender to give a tenfold dilution [13,14]. One gram of each soil sample was aseptically dispersed into 9ml of sterile peptone water in a test tube. After thorough mixing, 1ml portion of the mixture was aseptically transferred to a separate test tube containing 9ml of peptone water. These were mixed with a sterile rod, after which tenfold serial dilution was made according to the method of [12] and labeled appropriately.

2.3 Cultivation, Isolation and Identification of Organisms

Pour plate method was used. One milliliter (1ml) of each sample was aseptically dispensed in 9mls of sterile peptone water. The resulting suspension was diluted serially to the sixth dilution (106). One milliliter (1ml) of the 1st, 3rd and 6th diluents from each sample was dispensed into sterile petri dishes in duplicate. Eighteen millilitres (18mls) of molten Nutrient agar, MacConkey agar, Salmonella Shigella agar, Thiosulphate citrate bile salt sucrose (TCBS) agar was cooled to 45°C and poured into the different plates. The plates were incubated at 37°C for 24 hours [15]. The number of bacterial colonies on the plates with distinct growth were counted after incubation, multiplied by the dilution factors, and recorded as colony forming units per milliliter (cfu/ml). The organisms were later identified bv colonial morpholoav biochemical tests as described by Cheesbrough [16].

2.4 Antibiotic Susceptibility Testing

The agar diffusion technique as described by Cheesbrough, [16] was used. Sensitivity disks conventional antibiotics containing like Augmentin (20 µg), Amoxicillin (10 μg), Ciprofloxacin (5 µg), Cotrimoxazole (30 µg), Gentamicin (10 µg) and Nitrofurantoin (300 µg) manufactured by Abtek Biological Ltd., England were used for sensitivity test. A sterile wire loop was used to touch 3 to 5 discrete colonies and emulsified in 4 ml of sterile nutrient broth. The turbidity was matched to that of a freshly prepared 0.5 McFarland standard in a good light. A sterile swab was used to inoculate the isolate on to Muller-Hinton agar by streaking over the surface of the medium in three directions rotating the plate approximately 60 degrees to ensure even distribution. Antibiotic discs were duly

placed on the inoculated plates after 3 - 5 minutes and incubated aerobically at 35° C. Both control and test plates were examined after overnight incubation to ensure the growths were confluent or near confluent. A ruler was used to measure the diameter of each zone of inhibition in millimeter.

2.5 Determination of Minimum Inhibitory Concentration (MIC)

This was carried out following the guidelines of National committee for Clinical Laboratory Standards (NCCLS), [17]. The inoculums for the MIC test were obtained from an overnight culture of the test organism. At least 3 to5 discrete colonies of the same morphology were picked with a sterile wire loop and transferred into a tube containing 5ml of Nutrient broth which was incubated at 35°C for two hours. The turbidity of the actively growing broth culture was adjusted to obtain turbidity comparable to that of 0.5 Minimum McFarland standard. Inhibitory Concentration was determined by micro-dilution method using twofold serial dilution method. A volume of 0.1ml of the standardized inoculums was inoculated into a series of 9 test wells that contain a standard amount of broth and twofold serial dilutions of the antibiotic being tested after reconstitution was made as indicated; 1:2, 1:4, 1:8, 1:16, 1:32, 1:64, 1:128, 1:256, 1:512. The tubes were incubated aerobically at 37°C for 18 to 24 hours with two control tubes for each test batch. The lowest concentration (highest dilution) of the specimen that produces no visible bacterial growth (no turbidity) when compared with the control tube was regarded as the MIC (in ug/ml).

3. RESULTS

3.1 Mean Viable Bacterial Counts

Mean viable bacterial counts were generally very high (Table 1). Counts were highest for contaminated soil and least for workers hands. For meat contact surfaces, mean bacterial count was highest for slaughter slab (4.24×10^{6} Cfu/ml) and least for workers' hands (1.17×10^{6} Cfu/ml). Mean bacterial count was higher in washing water compared to rinsing water (5.30×10^{6} Cfu/ml versus 3.10×10^{6} Cfu/ml). Also, mean bacterial count was higher in contaminated soil compared to control soil samples (6.13×10^{6} Cfu/ml versus 3.25×10^{6} Cfu/ml).

Item	Mean \pm Standard deviation (x 10 ⁶ Cfu/ml)
Workers' hands	1.17 ± 0.30
Slaughter slab	4.24 ± 0.49
Butchering tables	2.5 ± 0.45
Butchering knifes	1.79 ± 0.25
Meat	1.50 ± 0.30
Washing water	5.30 ± 0.33
Rinsing water	3.10 ± 0.25
Control soil	4.25 ± 0.52
Contaminated soil	6.13 ± 0.24

Table 1. Mean \pm Standard deviation of total viable bacterial counts from various items

3.2 Distribution and Prevalence of the Isolated Bacteria

Before the distribution and prevalence of the isolated bacteria from different items in the abattoir is presented in Table 2. Escherichia coli had the highest prevalence (18.00%) with highest concentration in contaminated soil (3.10%), it was followed by Staphylococcus aureus, (this was confirmed by gram stain, catalase and coagulase tests) with total prevalence of 17.40% and highest concentration on slaughter slab (3.72%). Third most prevalent bacterium was Enterococcus species (16.15%), which also had highest concentration on slaughter slabs. Vibrio cholera (this was further confirmed by plating out on TCBS agar) recorded the least prevalence (0.62%) as it was isolated only from washing water. These organisms were confirmed using the respective biochemical tests.

3.3 Antimicrobial Susceptibility Test Result

Fig. 1 shows the zones of inhibition and interpretations of disc diffusion tests of isolated Gram-positive bacteria from meat samples. Marked resistance to the antibiotics used was observed.

Staphylococcus aureus was sensitive to gentamycin, ciprofloxacin and ceftriaxone, while *Enterococcus faecalis* showed sensitivity to gentamycin alone with intermediate sensitivity to ciprofloxacin, erythromycin and ceftriaxone.

There was also a marked resistance among Gram-positive organisms isolated from washing water samples (Fig. 2). Out of the ten antibiotics used, *Staphylococcus aureus* was sensitive to only three (gentamycin, ciprofloxacin and ceftriaxone), *Streptococcus spp* was sensitive to only cefuroxime, with intermediate sensitivity to

ceftriaxone and erythromycin. *Enterococcus* faecalis showed sensitivity to gentamycin, with intermediate sensitivity to ciprofloxacin.

High resistance to antibiotics was also recorded among Gram-negative isolates from meat samples (Fig. 3). *Escherichia coli* were sensitive to ciprofloxacin and penicillin, with intermediate sensitivity to pefloxacin and gentamycin. *Salmonella typhi* showed intermediate sensitivity to ciprofloxacin and ofloxacin, while *Shigella spp* showed intermediate sensitivity to ciprofloxacin.

Antibiotic resistance was also high for the Gramnegative isolates from washing water samples (Fig. 4). Escherichia coli were sensitive to only gentamycin, with intermediate sensitivity to streptomycin, ceporex and ofloxacin. Salmonella typhi was sensitive to only ciprofloxacin and intermediate to ofloxacin. Shigella spp were sensitive to gentamycin alone, while Vibrio cholerae were sensitive to gentamycin with intermediate sensitivity to ciprofloxacin. Proteus spp showed sensitivity to gentamycin and sensitivity to intermediate pefloxacin and ofloxacin.

The results of minimum inhibitory concentration (MIC), of selected sensitive antibiotics for both Gram-positive and Gram-negative bacteria isolated from meat and washing water samples are presented in Tables 3-10.

3.3.1 Gentamycin

In meat samples, *Salmonella* and *Shigella* species have the highest values (25µg/ml) of MIC while *Enterococcus feacalis* and *Proteus spp.* have the least values (3.13µg/ml). The highest MIC value in washing water samples was seen in *Pseudomonas aeruginosa* (100 µg/ml) while *Escherichia coli, Staphylococcus aureus* and *Enterococcus feacalis* have the least MIC values (3.13 µg/ml).

Isolates [°]	Tables	Meat	Workers hands	Butchering knives	Slaughter slab	Washing water	Rinsing water	Contaminated soil	Control	Total Prevalence
Escherichia coli	3(1.86%)	3(1.86%)	3(1.86%)	4(2.48%)	4(2.48%)	3(1.86%)	2(1.24%)	5(3.10%)	2(1.24%)	29(18.00%)
Pseudomonas aeruginosa	1(0.62%)	0(0.00%)	0(0.00%)	1(0.62%)	2(1.24%)	1(0.62%)	0(0.00%)	3(1.86%)	2(1.24%)	10(6.20%)
Salmonella typhi.	2(1.24%)	3(1.86%)	0(0.00%)	2(1.24%)	2(1.24%)	3(1.86%)	2(1.24%)	3(1.86%)	2(1.24%)	22(13.66%)
Shigella spp.	1(0.62%)	1(0.62%)	0(0.00%)	1(0.62%)	3(1.86%)	2(1.24%)	0(0.00%)	1(0.62%)	1(0.62%)	10(6.20%)
Staphylococcus	3(1.86%)	3(1.86%)	2(1.24%)	3(1.86%)	6(3.72%)	4(2.48%)	1(0.62%)	4(2.48%)	2(1.24%)	28(17.40%)
aureus	· · ·		· · · ·	, , , , , , , , , , , , , , , , , , ,	, , , , , , , , , , , , , , , , , , ,	х <i>у</i>	. ,	х <i>у</i>	· · · ·	, , , , , , , , , , , , , , , , , , ,
Klebsiella spp.	1(0.62%)	0(0.00%)	0(0.00%)	0(0.00%)	3(1.86%)	1(0.62%)	0(0.00%)	2(1.24%)	0(0.00%)	7(4.34%)
Vibrio cholerae	0(0.00%)	0(0.00%)	0(0.00%)	0(0.00%)	0(0.00%)	1(0.62%)	0(0.00%)	0(0.00%)	0(0.00%)	1(0.62%)
Streptococcus	1(0.62%)	0(0.00%)	0(0.00%)	0(0.00%)	2(1.24%)	1(0.62%)	0(0.00%)	1(0.62%)	1(0.62%)	6(3.72%)
spp										
Enterococcus	3(1.86%)	2(1.24%)	2(1.24%)	2(1.24%)	6(3.72%)	3(1.86%)	2(1.24%)	5(3.10%)	1(0.62%)	26(16.15%)
Feacalis										
Proteus spp.	2(1.24%)	2(1.24%)	2(1.24%)	3(1.86%)	4(2.48%)	3(1.86%)	2(1.24%)	3(1.86%)	1(0.62%)	22(13.66%)
Total	17(10.54%)	14(8.68%)	9(5.58%)	16(9.92%)	35(21.70%)	22(13.64%)	9(5.58%)	27(16.82%)	12(7.44%)	161(99.95%)

Table 2. Distribution and prevalence of isolates from different items

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Fig. 1. Zones of inhibition (mm) for Gram positive bacterial isolates from meat samples Key: Pef.= Pefloxacin, Gent.= Gentamycin, Cipro.= Ciprofloxacin, Sep.= Septrin,Strept.= Streptomycin, Ery.= Erythromycin, Roce.= Rocephine, Amox.= Amoxacillin, Zinna.= Zinnacef Ery.= Erythromycin, Roce.= Rocephine, Amox.= Amoxacillin, Zinna.= Zinnacef .



Fig. 2. Zones of inhibition (mm) for Gram positive bacterial isolates from washing water samples

Key: Pef.= Pefloxacin, Gent.= Gentamycin, Cipro.= Ciprofloxacin, Sep.= Septrin, Strept.= Streptomycin, Ery.= Erythromycin, Roce.= Rocephine, Amox.= Amoxacillin, Zinna.= Zinnacef .



Fig. 3. Zones of inhibition (mm) for Gram-negative bacterial isolates from meat samples Key: Pef.= Pefloxacin, Gen.= Gentamycin, Cipro.= Ciprofloxacin, Sep.= Septrin, Strepto.= Streptomycin, Amp= Ampicillin, CEP. Ceporex. OFX.= Ofloxacin, NA.= Nalidixic acid



Fig. 4. Zones of inhibition (mm) for Gram-negative bacterial isolates from washing water samples

Key: Pef.= Pefloxacin, Gen.= Gentamycin, Cipro.= Ciprofloxacin, Sep.= Septrin, Strepto.= Streptomycin, Amp= Ampicillin, CEP. Ceporex. OFX.= Ofloxacin, NA.= Nalidixic acid .

3.3.2 Ciprofloxacin

The highest MIC value in meat sample was observed in *Salmomella typhi, Shigella spp.* and *Enterococcus feacalis* (2.5 μ g/ml), while the least values was observed in *E. coli, Staphylococcus. aureus* and *Proteus spp.* (1.25 μ g/ml). The highest MIC value in washing water sample was seen in *Pseudomonas aeruginosa* (10 μ g/ml), while *Salmonella typhi* and *Staphylococcus. aureus* have the least values (1.25 μ g/ml).

3.3.3 Ampicillin

The highest MIC value in meat sample was observed in Salmonella and Shigella (62.50

µg/ml) while *E.coli* had the least MIC value (7.81µg/ml). In washing water sample, the highest MIC value was seen in *Pseudomonas* (125µg/ml) and least in *E. coli, Salmonella* and *Vibrio cholerae* (31.25µg/ml).

3.3.4 Ceftriaxone

The highest MIC value in meat samples was seen in *Enterococcus. feacalis* (31.25 μ g/ml), followed by *Streptococcus Spp.* (15.25 μ g/ml) while the least was observed in *Staphylococcus aureus* (7.81 μ g/ml). In washing water sample, the highest MIC was observed in *Enterococcus feacalis* (31.25 μ g/ml) and the least was in *Staphylococcus aureus* (3.91 μ g/ml).

Table 3. Minimum inhibitory concentrations (MICs) of Gentamycin (µg/ml) for bacterial isolates from meat samples

Isolates	800	400	200	100	50	25	12.50	6.25	3.13	1.56
Escherichia coli	-	-	-	-	-	-	-	-	+	+
Salmonella spp	-	-	-	-	-	-	+	+	+	+
Shigella spp	-	-	-	-	-	-	+	+	+	+
Staphylococcus aureus	-	-	-	-	-	-	-	-	+	+
Enterococcus feacalis	-	-	-	-	-	-	-	-	-	+
Proteus spp	-	-	-	-	-	-	-	-	-	+

Keys: Growth = + No growth = -

Table 4. Minimum inhibitory concentrations (MICs) of Ciprofloxacin (µg/ml) for bacterial isolates from meat samples

Isolates	40	20	10	5	2.5	1.25	0.63	0.31	0.16	0.08
Escherichia coli	-	-	-	-	-	-	+	+	+	+
Salmonella spp	-	-	-	-	-	+	+	+	+	+
Shigella spp	-	-	-	-	-	+	+	+	+	+
Staphylococcus aureus	-	-	-	-	-	-	+	+	+	+
Enterococcus feacalis	-	-	-	-	-	+	+	+	+	+
Proteus spp	-	-	-	-	-	-	+	+	+	+

Keys: Growth = + No growth = -

Isolates	500	250	125	62.50	31.25	15.25	7.81	3.91	1.95	0.98
Escherichia coli	-	-	-	-	-	-	-	+	+	+
Salmonella spp	-	-	-	-	+	+	+	+	+	+
Shigella spp	-	-	-	-	+	+	+	+	+	+
Proteus spp	-	-	-	-	-	+	+	+	+	+

Table 5. Minimum inhibitory concentrations (MICs) of Ampicillin (µg/ml) for Bacterial isolates from meat samples

Keys : Growth = + No growth = -

Table 6. Minimum inhibitory concentrations (MICs) of Ceftriazone (µg/ml) for bacterial isolates from meat samples

Isolates	1000	500	250	125	62.50	31.25	15.25	7.81	3.91	1.95
Staphylococcus aureus	-	-	-	-	-	-	-	-	+	+
Enterococcus feacalis	-	-	-	-	-	-	+	+	+	+
		14	0							

Keys: Growth = + No growth =

Table 7. Minimum inhibitory concentrations (MICs) of Gentamycin (µg/ml) for bacterial isolated from washing water samples

Isolates	800	400	200	100	50	25	12.50	6.25	3.13	1.56
Escherichia coli	-	-	-	-	-	-	-	-	-	+
Pseudomonas aeruginosa	-	-	-	-	+	+	+	+	+	+
Salmonella spp	-	-	-	-	-	-	+	+	+	+
Shigella spp	-	-	-	-	-	-	-	+	+	+
Staphylococcus aureus	-	-	-	-	-	-	-	-	-	+
Klebsiella spp	-	-	-	-	-	-	-	-	+	+
Vibrio cholerae	-	-	-	-	-	+	+	+	+	+
Streptococcus spp	-	-	-	-	-	+	+	+	+	+
Enterococcus feacalis	-	-	-	-	-	-	-	-	-	+
Proteus spp	-	-	-	-	-	-	-	-	+	+

Keys: Growth = + No growth = -

Table 8. Minimum inhibitory concentrations (MICs) of Ciprofloxacin (µg/ml) for bacterial isolated isolates from washing water samples

40	20	10	05	2.50	1.25	0.63	0.31	0.16	0.08
-	-	-	-	-	+	+	+	+	+
sa-	-	-	+	+	+	+	+	+	+
-	-	-	-	-	-	+	+	+	+
-	-	-	-	+	+	+	+	+	+
-	-	-	-	-	-	+	+	+	+
-	-	-	-	-	+	+	+	+	+
-	-	-	-	-	+	+	+	+	+
-	-	-	-	-	+	+	+	+	+
-	-	-	-	-	+	+	+	+	+
-	-	-	-	+	+	+	+	+	+
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Keys: Growth = + No growth = -

4. DISCUSSION

It was observed during this study that the abattoir had rather, very dirty environment. It was also of slaughter slab type, with poor sanitary conditions. The study was conducted using a total of 70 samples, ten for each of slaughter slabs, butchering tables, workers' hands, butchering knives, effluent water, meat and soil, from the abattoir. The mean viable colony counts were generally high (Table 1), similar to the findings of Onuoha *et al.*, [18] in the same abattoir and with

most of the other studies cited in this work. This emphasizes the fact that waste water from the abattoirs may be a source of spread of bacteria of public health importance, and the need for adequate treatment of waste water before discharge to the environment.

There was higher bacterial count in washing section, compared to rinsing. This is similar to the findings by Neboh *et al.* [19] in Ijebu-Igbo, Ogun State Nigeria. The findings also corroborate the previous suggestion by Nwanta and his colleagues [20], that our abattoirs, meat processing plants, and meat inspection services were poor. These are of huge public health importance.

Very high mean colony count from soil samples contaminated by abattoir waste and effluent water was revealed. Similar findings have been recorded in a previous study conducted by Mbajiuka and his colleagues [21], in Umuahia. This study also revealed much higher mean colony count for contaminated soil compared to control soil samples. This agrees with the findings by Mbajiuka *et al.*, [21] which suggested that high microbial load in abattoir wastes had a negative effect on the microbial population in the soil.

A look at the meat contact surfaces showed highest value of mean colony count for slaughter slab, compared to workers' hands, butchering tables and knives. Similar finding was recorded by Onuoha *et al.*, [22] in Ebonyi State. This study, however, included two more contact surfaces – workers' hands and butchering knives – with mean colony count still remaining highest for slaughter slab. It therefore remains pronounced that the slaughter slabs in the abattoirs should receive more sanitary attention.

Frequency of occurrence of isolated bacteria was also evaluated and *Escherichia coli* had the highest prevalence. This bacterium has been incriminated in cases of stomach ache and diarrhea and is suggestive of high level of contamination of abattoir environments with fecal materials. Of interest is the isolation of *Vibrio cholerae* from a washing water sample from the abattoir. Adesemoye *et al.*, [12] previously isolated *Vibrio spp* from waste water sample from Agege and Ojo areas of Lagos State. This raises concern that the abattoirs can serve as sources of cholera outbreak. Therefore, proper cooking of meat and adequate treatment of abattoir wastes before disposal are very important.

Escherichia coli. Salmonella tvphi and Staphylococcus aureus constituted the organisms that had highest frequency of occurrence in meat samples. Adesiji et al., [23] reported S. aureus to be the highest occurring organism in beef and other retail raw meat in Oshogbo, Nigeria. High prevalence of salmonella typhi in meat samples in this study goes to draw our attention to the fact that poorly cooked meat from these abattoirs can expose the public to outbreak of typhoid enteritis. Enterococcus fecalis also had a significant frequency of occurrence in meat samples in this study; corroborating the assertion that there is high contamination of meat samples from our abattoirs with fecal matter.

Table 9. Minimum inhibitory concentrations (MICs) of Ampicillin (µg/ml) for bacterial isolates
from washing water samples

Isolates	500	250	125	62.50	31.25	15.25	7.81	3.91	1.95	0.98
Escherichia coli	-	-	-	-	-	+	+	+	+	+
Pseudomonas aeruginosa	-	-	-	+	+	+	+	+	+	+
Salmonella spp	-	-	-	-	-	+	+	+	+	+
Shigella spp	-	-	-	-	+	+	+	+	+	+
Klebsiella spp	-	-	-	-	+	+	+	+	+	+
Vibrio cholerae	-	-	-	-	-	+	+	+	+	+
Proteus spp	-	-	-	-	+	+	+	+	+	+

Keys : Growth = + No growth = -

Table 10. Minimum inhibitory concentrations (MICs) of Ceftriazone (µg/ml) for bacterial isolates from washing water samples

Isolates	1000	500	250	125	62.50	31.25	15.25	7.81	3.91	1.95
Staphylococcus aureus	-	-	-	-	-	-	-	-	-	+
Streptococcus spp.	-	-	-	-	-	-	-	+	+	+
Enterococcus feacalis	-	-	-	-	-	-	+	+	+	+
		14	^							

Keys: Growth = + No growth = -

Escherichia coli. Salmonella tvphi. Staphylococcus aureus, Enterococcus fecalis and Proteus spp. had equal highest frequency of occurrence in waste water samples in this study. Shigella spp. had second highest frequency of occurrence, followed equally by Pseudomonas aeruginosa, Klebsiella spp., Vibrio cholerae and Streptococcus spp. This picture goes to bring back what Onuoha et al., [22] have already found in their study, which they conducted in Ebonyi State, that waste water samples in their abattoir had high concentration of contaminants and bacteria of mostly enteric origin - fecal contamination.

There is also high prevalence of organisms of enteric origin in soil samples contaminated with the abattoir waste, with highest frequency occurring with Escherichia coli and Enterococcus fecalis. This is due to indiscriminate dropping of intestinal contents of slaughtered animals on abattoir soil. Other isolated organisms were Staphylococcus aureus. Pseudomonas aeruginosa, Salmonella typhi, Proteus spp, Klebsiella spp, Shigella spp and Streptococcus spp. This is similar to the results got by Onuoha et al., [22] Nwachukwu et al., [24] and others. They are of the common opinion, just like in this study, that high microbial load in abattoir wastes had negative effects on the microbial population of soil. Escherichia coli, Pseudomonas aeruainosa. Salmonella tvphi and Staphylococcus aureus isolated from meat contact surfaces in this study are identical to those isolated by Onuoha et al., [18]. However, other bacteria, Shigella spp, Klesiella spp, Streptococcus spp, Enterococcus faecalis and *Proteus spp* – are isolated from contact surfaces in this present study. This may have resulted from environmental variations. Furthermore, in addition to slaughter slab and tables, which they used as their contact surfaces, this study included butchering knives and workers' hands.

The presence of multi-drug-resistant strains of isolated bacteria in this study environment could act as means of transferring antibiotic resistance to other bacteria, which is also a potential public health challenge. Values of minimum inhibitory concentration, in this study, being high for *Pseudomonas aeruginosa* reflects the fact already emphasized by Seol *et al.*, [25] that it is notorious for its resistance to antibiotics and therefore being a particularly dreaded pathogen. A study conducted in Ibadan by Rabiu and his colleague [26] showed that *Pseudomonas*

species isolated from waste water of Akinvele abattoir. demonstrated multi-drug resistant pattern, showing that abattoir wastewater could serve as important vehicle for sustenance of multi-drug resistant bacteria in aquatic ecosystem and transmission of multi-drug resistant disease causing bacteria to humans. Hence there is need to ensure adequate treatment of abattoir wastewater to degrade bacteria population, especially the potential pathogenic strains, before they are eventually released into the environment.

This present study also revealed high MIC values for Enterococcus spp and Streptococcus spp. Studies have shown that resistance of Streptococci and Enterococci to different classes of antimicrobials is increasing worldwide [27,28]. This drug resistant pattern by Enterococcus has been demonstrated in Imo State in a study using urine and high vaginal swab samples [29]. In Egypt, high prevalence of *E. faecalis* have been documented in cases of urinary tract infection, with 100 percent multi-drug-resistance rate, indicating a serious problem in treating infections by this organism [30]. By the 2000's, high level resistance (MIC \geq 2000µg/ml), was seen for enterococcus spp to gentamycin and other aminoglycosides [31]. In their study which involved collection of clinical samples from different colleges and hospitals, in Bhopal India, they found multi-drug-resistant Enterococci, which was a threat to patients' safety. Also in Iran, a study has shown that multi-antibiotic resistance to conventional antibiotics, by Enterococci, was significantly high [32].

5. CONCLUSION

The findings in this study, in agreement with those of other similar studies, have shown high bacterial contamination of our abattoir environments, water and meat. The abattoir environments are rather terribly dirty. Slaughter slabs are associated with high bacterial contamination. Isolated bacteria have shown commonly, alarming rate of resistance to antibiotics; this poses great challenge to public health, as it may serve as source of transfer of drug resistance to other bacteria and into the wider environment.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

CONSENT

As per international standard or university standard, Participants' written consent has been collected and preserved by the author(s).

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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