

Tropical Journal of Phytochemistry & Pharmaceutical SciencesAvailable online at <https://www.tjpps.org>**Original Research Article****Bioburden and Antibacterial Susceptibility Pattern of Hand Manual Grinders used in different Markets in Ozoro, Delta State**Nkeonyeasua J. Ehiwario^{1*}, Emmanuel O. Eromosele², Joshua O. Orogu¹, Uche B. Okobia¹ and Ejiro Idollo³¹Department of Microbiology, Delta State University of Science and Technology, Ozoro²Department of Medical Biochemistry, Faculty of Basic Medical Sciences, Ambrose Alli University, Ekpoma, Edo state.³Department of Science Education (Integrated Science), Delta State University, Abraka.

ABSTRACT

In Nigeria, the situation of most markets, experiences poor environmental sanitation and personal hygiene thereby encouraging the spread of pathogens and infectious diseases in the communities. This study examined the bioburden and antibacterial susceptibility pattern of hand manual Grinders used in different markets in Ozoro, Delta State. Twenty (20) samples (5 samples from each category of food items: Pepper/Spices, Crayfish, Ogbono and Egusi) were randomly collected by aseptically scrapping the market hand manual grinders initially used to ground the selected food stuffs. The bacterial mean total viable count (cfu/g) for sample A (Pepper/Spices) is 2.1×10^5 cfu/g, sample B (Crayfish) 2.6×10^5 cfu/g, sample C (Ogbono) 3.1×10^5 cfu/g and sample D (Egusi) 3.5×10^5 cfu/g. A total of 62 bacterial species comprising of Seven (7) genera was identified which includes *Staphylococcus aureus*, *Bacillus* species, *Escherichia coli*, *Enterobacter aerogenes*, *Pseudomonas aeruginosa*, *Salmonella typhi* and *Streptococcus* species. *Staphylococcus aureus* has the highest prevalence of occurrence 24(38.7%) while the least was recorded by *Salmonella typhi* 3(4.8%) and *Streptococcus* species 3(4.8%). The susceptibility testing of the bacteria isolates against different antibiotics indicates that Ciprofloxacin, Levofloxacin, Gentamicin and Ofloxacin are the most effective against the bacteria tested while the least was Augmentin and Amoxicillin. The resistance of the bacterial isolates to Augmentin and Amoxicillin in this present study is of public health concern. As such, hand manual grinders used in the market for grinding food stuffs should be properly cleaned before and after use, market sanitation and personal hygiene should be observed and encouraged regularly to reduce spread of infectious diseases.

Keywords: Bioburden, Grinders, Pepper/spices, Crayfish, Ogbono, Egusi, Market, Bacterial isolates, Antibiotics and Susceptibility

Received 24 December 2024

Revised 21 February 2025

Accepted 28 February 2025

Published online 04 March 2025

Copyright: © 2025 Ehiwario *et al.* This is an open-access article distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.**Introduction**

A market is a setting where different categories of people gather regularly for the purpose of purchasing and sales of goods and services. In Nigeria, the situation of most markets experiences poor environmental sanitation and poor personal hygiene thereby encouraging the spread of pathogens and infectious diseases. Government agencies saddled with the responsibility of environmental sanitation, stakeholders and individuals making use of the market should be involved in improving sanitation as it has significant beneficial impact on the health of both in households and across communities. Markets contribute significantly to and strengthen the economic life of the people and give a large measure of economic opportunities thereby helping to sustain livelihood. Markets are one of the most focal points for the interaction of the people because almost everyone has to visit the market for one service or the other.

*Corresponding author. Email: ehiwariojulius@gmail.com

Tel: +2347058151733

Citation: Ehiwario NJ, Eromosele EO, Orogu JO, Okobia UB, Idollo E. Bioburden and Antibacterial Susceptibility Pattern of Hand Manual Grinders used in different Markets in Ozoro, Delta State. Trop J Phytochem Pharm. Sci.4 (2) 85 – 91 <http://www.doi.org/10.26538/tjpps/v4i2.9>

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria.

They are responsible for most of the spatial interactions that take place in a city. Thus, any epidemic that emanates from the market will inevitably affect the whole population.¹The biggest cause of poor sanitation globally is simply lack of education. Even the simple act of washing hands regularly can have a huge impact on the overall health of a community. To address the enormous problems of environmental sanitation in Nigeria, the Federal Ministry of Environment (FMOE) through the National Environmental Sanitation Policy, identified market and abattoir sanitations as areas of concern. This was sequel to the overwhelming sanitation problems in markets and abattoirs that includes improper refuse disposal, inadequate water supply, and as well as overcrowding and exposure of food and meat to flies, rodents and contaminants.² According to,³ 23.5% of the population defecate and urinate in the open” as result of gross inadequacy of sanitary facilities. The essential components of environmental sanitation include: solid waste management; medical waste management; excreta and sewage management; food sanitation; sanitary inspection of premises; market and abattoir sanitation; adequate potable water supply; school sanitation; pest and vector control; management of urban drainage; control of reared and stray animals; disposal of the dead animals; weed and vegetation control; hygiene education and promotion.⁴ Microbial contamination of food is a major public health problem since it affects millions of people worldwide. An unhygienic practice that occurs within the food processing environment brings

about contamination with pathogens, turning food into a risk for the safety of the consumers.⁵

As reported by,⁶ in their investigation through weeks of daily monitoring of the level of hygiene outcome in a local community market in Rivers State, Nigeria and they revealed that cups used in the measurement of products such as rice, beans, melon and other seed-like products are not cleaned before and after use overtime, thus this would likely promote the presence and proliferation of microorganisms in these cups and measuring buckets, and subsequent transfer same to the products being measured. When these seed products are taken to be ground, the person concerned with this also, does not take out time to clean his/her machine before and after use, thus providing another good environment for the growth and possible colonization of new microorganisms different from those found in the seeds. Nonetheless, blenders are thus, machines (either electrically or manually operated) used to cut or crush substances (especially food and fruits) into smaller pieces.

In most rural communities and markets, after blending,⁶ observed that the blenders are not washed. But are used over and over again, which often provides conducive media for the growth of microorganisms, possibly pathogenic ones to thrive. Nonetheless, such an unprofessional practice remains a massive promoter of an outbreak of foodborne epidemic, such as gastroenteritis and other seeming stomach disorders among unsuspected subjects, especially in rural settings, where access to the functional health system and availability of drugs has been a massive problem begging for attention over the years.⁷

Recent studies have shown the presence of bacterial pathogens on hard, nonporous surfaces such as kitchen surfaces, floor surfaces, toilet surfaces, toilet door handles, car door handles etc.,^{8, 9, 10} from which pathogens are easily transmitted to unsuspecting members of the public posing a potential risk to vulnerable, immune-compromised individuals.^{11, 8} Currently, some of these bacterial pathogens have become antibiotic resistant, which is a major public health crisis facing the world today.^{12, 13} Therefore there is an urgent need to improve standards hygienic use of hand manual grinders used for grinding food stuffs like pepper/spices, crayfish, ogbono and egusi in the markets in order to reduce the spread of infectious diseases. Hence the aim of this research was to isolate and identify bacterial species associated with hand manual grinders used for grinding food stuffs in the markets and further carry out antibiotic sensitivity study on the isolates.

Materials and Methods

Description of Study Area

This research was conducted in Ozoro, Isoko North L.G.A of Delta state in Nigeria. Ozoro is between latitude 5° 32' 30''N and longitude 6° 13' 38''E situated along the high way between Ughelli and Kwale towns. Ozoro is the headquarters of the Isoko North Local Government Area, one of the two administrative units in the Isoko region of Delta State, Southern Nigeria. Ozoro had been incorrectly spelt "Usoro" in some older maps of Nigeria. It is densely populated and their major occupation is farming and trading. Ozoro community climate is characterized by two seasons: the wet and dry seasons. This location was chosen because it is a fast growing community in Isoko with high social activities being boosted by the presence of markets: the Ozoro Big market, which is scheduled for the 4th day of every week; Small market and Akporio market which are daily markets and tertiary institution (Delta State University of Science and Technology, Ozoro).

Informed Consent

Unannounced visits were made to the markets (Ozoro Big market, Small market and Akporio market) in Ozoro, Isoko North Local Government Area of Delta State. Consent was sought and obtained verbally from the owners of the hand manual grinders in the different markets in Ozoro and with the letter of introduction from the Department of Microbiology, Delta State University of Science and Technology, Ozoro.

Collection of Samples

The samples used in this research were grouped into four (4) categories based on the hand manual grinder used for grinding different food stuff items which are pepper/spices, crayfish, Ogbono and Egusi; this was to ensure that representative samples was evenly collected. A total of twenty (20) samples (5 samples from each categories of food items) were randomly collected among the four categories of samples by aseptically scrapping the market hand manual grinders that have been used to ground selected food stuff samples and was properly labeled as A₁, A₂, A₃, A₄, and A₅ for Pepper/Spices samples; B₁, B₂, B₃, B₄ and B₅ for Crayfish samples; C₁, C₂, C₃, C₄ and C₅ for Ogbono samples and D₁, D₂, D₃, D₄ and D₅ for Egusi samples before they were aseptically delivered into sterile containers and was immediately transported to the laboratory in an ice-chest for bacteriological analysis as described by.^{14, 6}

Bacteriological Analysis

Isolation of Bacteria species in the Samples

One gram (1g) of each samples (pepper/spices, crayfish, ogbono and egusi) was added to 9ml of normal saline from which a 10-fold serial dilution was carried out and 0.1 ml from dilution (10⁻³) of each sample was plated on freshly prepared Nutrient agar plates in duplicates; these were incubated in a well regulated incubator for 24 hours at 37°C for the isolation of bacteria species associated with the samples.^{14, 6} Pure cultures were obtained by sub-culturing distinct colonies on freshly prepared culture plates.

Identification of Bacteria Isolates

Identification of bacterial isolates was carried out using morphological characteristics (Gram staining, cell morphology and cell arrangement), and biochemical tests (motility test, indole test, urease test, citrate test, coagulase test, catalase test and oxidase test) were carried out to identify and characterize the bacteria isolates. Pure cultures that had been identified were stored on nutrient agar slants for further antibiotic study as described in.^{14, 6}

Antibacterial Susceptibility Testing

Antibacterial susceptibility testing was carried out on the bacterial isolates by using Modified Kirby Bauer Disk Diffusion method using Mueller-Hinton agar (MHA). It was performed according to the Clinical and Laboratory Standards Institute (CLSI) guidelines¹⁵. 0.5 McFarland turbidity standards was used to compare the turbidity of the bacterial suspension by transferring three to five colonies of the isolated bacteria to a tube containing 5 ml normal saline and mixed gently until a homogenous suspension formed. The bacterial suspension was then swabbed over the entire surface of Mueller-Hinton agar using a sterile cotton swab. Antibiotic discs were applied within fifteen minutes using sterile forceps on the surface of medium and incubated at 37 °C for 18 to 24 hours. The zone of inhibition of growth around each disk was then measured in millimeters. The antibiotic discs tested for isolate of bacterial species are Ciprofloxacin (5µg), Augmentin (30µg), Gentamycin (10µg), Levofloxacin (5µg), Streptomycin (30µg), Ofloxacin (5µg), Imipenem (5µg), Amoxicillin (30µg) and Setrin (30µg). The results were expressed in percentage of susceptibility/resistance according to the absence or presence/diameter of inhibition: - = No zone of inhibition (Resistant); + = 5mm zone of inhibition (low resistance), ++ = 10 to 14mm (intermediate), +++ = 15 to 17mm zone of inhibition (sensitive).^{16, 10}

Statistical Analysis

Statistical analysis was carried out using Microsoft Excel (version 2007) and was presented as simple percentages.

Enterobacter aerogenes, *Pseudomonas aeruginosa*, *Salmonella typhi* and *Streptococcus* species as indicated in Table 2.

Results and Discussion

The bacteriological analysis of bacterial species associated with hand manual grinders used for grinding food stuffs like pepper, spices, crayfish, ogbono and egusi in different markets in Ozoro, Delta State, results show that the grinders harbor bacteria loads. The environmental conditions of most markets in developing countries are far below the required hygienic practices. According to,¹⁷ maintaining proper sanitation and behavior of food handlers is crucial in determining the quality of food. Most of the hand manual grinders used for grinding different food stuff items like pepper, spices, crayfish, ogbono and egusi are not cleaned before and after been used and this tends to be a breeding medium for the growth of microorganisms. According to,⁶ probably due to the large number of persons coming to grind food stuffs, the persons doing the grinding end up re-using the grinders without proper cleaning not minding the source of the food stuff. Sometimes, the food stuff may be already or almost spoilt (as in the case of some fresh pepper and tomato brought for blending), thus depositing some food spoilage microorganisms on the blender. Without proper cleaning, these organisms may replicate sporadically and be transferred to the new food stuff brought for grinding also. These scenario tend to put the unsuspecting public at massive potential risk with strong public health concern of food poisoning outcome, if not checked in good time to nip it at the board.⁶ Table 1 revealed the bacterial total viable count (cfu/g) of the samples examined. The sample D (Egusi) has the highest mean count of 3.5×10^5 cfu/g followed by sample C (Ogbono) with (3.1×10^5) cfu/g, sample B (Crayfish) having (2.6×10^5) cfu/g while sample A (Pepper/Spices) has the least mean count of 2.1×10^5 cfu/g. The bacterial total viable count (cfu/g) for sample A (Pepper/Spices) ranges from 1.4×10^5 cfu/g to 3.0×10^5 cfu/g with a mean count of 2.1×10^5 cfu/g, sample B (Crayfish) 1.8×10^5 cfu/g to 3.2×10^5 cfu/g with a mean count of 2.6×10^5 cfu/g, sample C (Ogbono) 2.8×10^5 cfu/g to 3.5×10^5 cfu/g with a mean count of 3.1×10^5 cfu/g, sample D (Egusi) 2.8×10^5 cfu/g to 4.0×10^5 cfu/g with a mean count of 3.5×10^5 cfu/g. The sample D (Egusi) has the highest mean count of 3.5×10^5 cfu/g and this is not in line with the research findings of,⁶ who reported that the blender used to ground Ogbono had the highest bacterial count of 1.67×10^6 compared with that of Egusi (2.80×10^4). This could be as a result of higher moisture content of Egusi compared to other food stuffs examined and high consumption of Egusi in the study area (Ozoro). Sample A (pepper/spices) has the least mean count of 2.1×10^5 cfu/g and this may be as a result of the low moisture content of the sample. In this present study, the hand manual grinders used for grinding different food stuff items were examined and a total of 62 bacterial species was obtained which accounted for 40 gram positive and 22 gram negative bacteria. Seven (7) bacteria species were isolated which includes *Staphylococcus aureus*, *Bacillus* species, *Escherichia coli*,

Table 1: Bacterial Total Viable Count (cfu/g)

Sample code for the food stuffs	Dilution	No of colonies	Bacterial count (cfu/g)	Bacterial Mean count
Pepper/Spices				
A ₁	10 ⁻³	230	2.3x10 ⁵	2.1x10 ⁵ cfu/g
A ₂	10 ⁻³	307	3.0x10 ⁵	
A ₃	10 ⁻³	201	2.0x10 ⁵	
A ₄	10 ⁻³	146	1.4x10 ⁵	
A ₅	10 ⁻³	200	2.0x10 ⁵	
Crayfish				
B ₁	10 ⁻³	322	3.2x10 ⁵	2.6x10 ⁵ cfu/g
B ₂	10 ⁻³	202	2.0x10 ⁵	
B ₃	10 ⁻³	290	2.9x10 ⁵	
B ₄	10 ⁻³	182	1.8x10 ⁵	
B ₅	10 ⁻³	300	3.0x10 ⁵	
Ogbono				
C ₁	10 ⁻³	330	3.3x10 ⁵	3.1x10 ⁵ cfu/g
C ₂	10 ⁻³	288	2.8x10 ⁵	
C ₃	10 ⁻³	350	3.5x10 ⁵	
C ₄	10 ⁻³	300	3.0x10 ⁵	
C ₅	10 ⁻³	293	2.9x10 ⁵	
Egusi				
D ₁	10 ⁻³	381	3.8x10 ⁵	3.5x10 ⁵
D ₂	10 ⁻³	400	4.0x10 ⁵	
D ₃	10 ⁻³	373	3.7x10 ⁵	
D ₄	10 ⁻³	289	2.8x10 ⁵	
D ₅	10 ⁻³	306	3.0x10 ⁵	

Table 2: Morphological characteristics and Biochemical tests

Gram Reaction	Cell Morphology	Cell Arrangement	Motility	Indole	Urease	Citrate	Coagulase	Catalase	Oxidase	Identified Organisms
+	Cocci	Cluster	-	-	-	-	+	+	-	<i>Staphylococcus aureus</i>
+	Rod	Chain	+	-	-	+	-	+	+	<i>Bacillus</i> species
-	Rod	Single	+	+	-	-	-	+	-	<i>Escherichia coli</i>
-	Rod	Chain	+	-	-	+	-	+	-	<i>Enterobacter aerogenes</i>
-	Rod	Single	+	-	-	+	-	+	+	<i>Pseudomonas aeruginosa</i>
-	Rod	Pair	+	-	-	-	-	+	-	<i>Salmonella typhi</i>
+	Cocci	Chain	-	-	-	-	-	-	-	<i>Streptococcus</i> species

This is in accordance with,⁶ who isolated similar organisms except *Enterobacter aerogenes*, *Salmonella typhi* and *Streptococcus* species.

In the report of,⁵ on microbiological assessment of cutlery, isolated *Staphylococcus aureus* and *Escherichia coli* which were also isolated in this present study. The presence of these microorganisms in the samples could be as a result of human interactions, cross contamination of food stuffs at points of processing and dust particles in the market which are bioburden. Table 3 shows the distribution of Bacterial isolates from the samples examined. *Staphylococcus aureus* has the highest prevalence of occurrence of 24(38.7%) followed by *Bacillus* species 13(21%), *Escherichia coli* 9(14.5%), *Enterobacter aerogenes* 6(9.7%), *Pseudomonas aeruginosa* 4(6.5%), *Salmonella typhi* 3(4.8%) and *Streptococcus* species 3(4.8%) respectively. Of the samples examined, Egusi sample has the highest prevalence of bacteria isolates of 27(43%) followed by Ogbono sample 16(26%) while the least is Pepper/Spices 6(10%) as shown in Figure 1. The frequency of bacterial isolates from the different food stuff samples examined revealed that Sample D (Egusi) has the highest bioburden followed by Sample C (Ogbono) while the least was Sample A (Pepper/spices) as indicated in Figure 2.

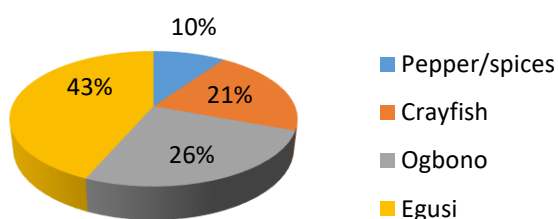


Figure 1: Prevalence of bacterial isolates according to food stuff items

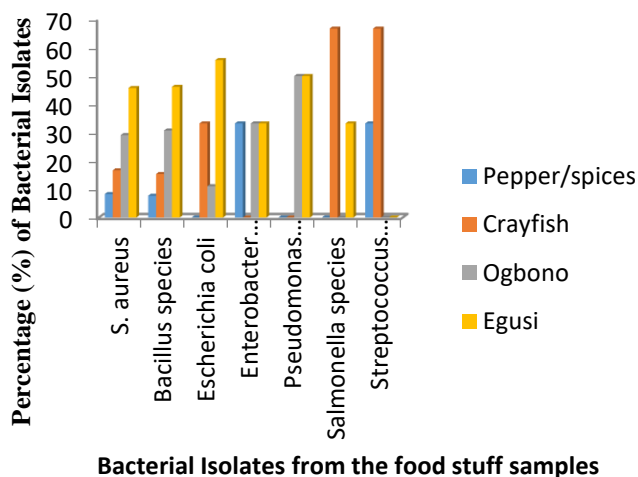


Figure 2: Frequency of Bacterial isolates from the different food stuff samples examined

Table 4 displays the distribution of isolates according to Gram's Reaction. A total of three (3) gram positive bacterial isolates with occurrence of 40(64.5%) while four gram negative bacterial isolates having 22(35.5%). For the gram positive bacteria, *Staphylococcus aureus* has the highest prevalence of 24(38.7%) while for the gram negative bacteria, *Escherichia coli* has the highest prevalence of 9(14.5%) as displayed in Table 4.

Table 3: Distribution of Bacterial isolates from the samples examined

Bacterial isolates	Prevalence of Isolates according to food stuff items				Total (%)
	A (%)	B (%)	C (%)	D (%)	
<i>Staphylococcus aureus</i>	2(8.3)	4(16.7)	7(29.2)	11(45.8)	24(38.7)
<i>Bacillus</i> species	1(7.7)	2(15.4)	4(30.8)	6(46.2)	13(21.0)
<i>Escherichia coli</i>	0(0)	3(33.3)	1(11.1)	5(55.6)	9(14.5)
<i>Enterobacter aerogenes</i>	2(33.3)	0(0)	2(33.3)	2(33.3)	6(9.7)
<i>Pseudomonas aeruginosa</i>	0(0)	0(0)	2(50)	2(50)	4(6.5)
<i>Salmonella typhi</i>	0(0)	2(66.7)	0(0)	1(33.3)	3(4.8)
<i>Streptococcus</i> species	1(33.3)	2(66.7)	0(0)	0(0)	3(4.8)
Total	6(10)	13(21)	16(26)	27(43)	62(100)

A = Pepper/Spices sample, B = Crayfish sample, C = Ogbono sample and D = Egusi sample.

Table 4: Distribution of Isolates according to Gram's

Isolates	Reaction
Gram Positive Bacteria	Number of Isolates (%)
<i>Staphylococcus aureus</i>	24(38.7)
<i>Bacillus</i> species	13(21.0)
<i>Streptococcus</i> species	3(4.8)
Sub-total	40(64.5)
Gram Negative Bacteria	
<i>Escherichia coli</i>	9(14.5)
<i>Enterobacter aerogenes</i>	6(9.7)
<i>Pseudomonas aeruginosa</i>	4(6.5)
<i>Salmonella typhi</i>	3(4.8)
Sub-total	22(35.5)
Total	62(100)

The result from this study shows that *Staphylococcus aureus* has the highest prevalence of 24(38.7%). This is in line with to the result of,⁶ who reported that *Staphylococcus aureus* had the highest frequency with a percentage occurrence of 12(24%) though lesser than the prevalence of *Staphylococcus aureus* in the present research. *Staphylococcus aureus* is gram positive bacteria which are cocci in shape, arranged in clusters (grape-like) and that are a major bacterial human pathogen which causes a wide variety of clinical manifestation.¹⁸ They are found on the skin, nostrils, mucous membranes and humans are the major reservoir host.^{19, 20} *S. aureus* does not normally cause infection on healthy skin; however, if it is allowed to enter the bloodstream or internal tissues, these bacteria may cause a variety of potentially serious infections.¹⁸ Transmission is typically from direct contact. However, some infections involve other transmission methods.²¹ Humans are the major reservoir host for *S.*

aureus.^{19, 20} It is estimated that up to half of all adults are colonized, and approximately 15% of the population persistently carry *S. aureus* in the anterior nares. Some populations tend to have higher rates of *S. aureus* colonization (up to 80%), such as health care workers, persons who use needles on a regular basis (ie, diabetics and intravenous drug users), hospitalized patients, and immune-compromised individuals. *S. aureus* can be transmitted person-to-person by direct contact or by fomites.^{18, 21, 22} *Staphylococcus aureus* are one of the most common bacterial infections in humans and are the causative agents of multiple human infections, including bacteremia, infective endocarditis, skin and soft tissue infections, osteomyelitis, pulmonary infections, gastroenteritis and urinary tract infections.²²

Bacillus species are gram positive, rod-like shape, cell arranged in chain, spore forming bacteria which are ubiquitous in nature. With their resistant endospores, they can survive in the harshest climatic and edaphic conditions. This could be the reason for them been the second highest in prevalence 13(21%) in this present study. The presence of *Bacillus* species in food stuffs samples examined could be as a result of sand or dust blown up by air current into the food stuffs before grinding or deposited on the hand manual grinders in the market being that soil is their natural habitat.

The isolation of *Bacillus* species with prevalence of 6(46.2%) from the hand manual grinder that was used to grind Ogbono is in accordance with research reports of,^{6, 23} who isolated the same organism and some other pathogens from blenders used in grinding some food stuffs in a local community market in Rivers State and fermented Ogbono seeds respectively.

Streptococcus species are gram positive cocci which are arranged in chains or pairs. In this research, *Salmonella typhi* and *Streptococcus* species has the least prevalence of 3(4.8%) each, but *Streptococcus* species was absent in sample C (Ogbono) and D (Egusi). *Streptococcus* species originate from several natural sources, including humans and many animal species, in which they naturally colonize the nasal passage, pharynx, and mucosal surfaces of the mouth and intestinal tract.²⁴ Poor hygiene leads to contamination of food during handling and preparation through coughing, sneezing, or from skin lesions, from people infected with human pathogens,²⁵ and as such prevention of such contamination is important and can be achieved by observing personal hygienic practices. *Streptococcus* species are responsible for many cases of endocarditis, meningitis, bacterial pneumonia, erysipelas, necrotizing fasciitis (the 'flesh-eating' bacterial infections) and streptococcal pharyngitis (strep throat).

The presence of enteric bacteria (bacteria that live and reside within the intestinal tract of animals and humans) pathogens such as

Salmonella typhi, *Enterobacter species* and *Escherichia coli* in this study, and some are pathogenic, causing disease and food poisoning in humans, can be picked up easily from objects that are fecally contaminated and their presence in this present research is indication of fecal contamination and poor personal hygiene by the users hand manual grinders. According to²⁶, *Salmonella typhi* is the major cause of typhoid fever which is a leading cause of disease and death in Nigeria today.

Pseudomonas aeruginosa occurred only in sample C (Ogbono) and D (Egusi) having a total prevalence of 4(6.5%). *Pseudomonas aeruginosa* is a non-spore forming gram-negative rod-shaped bacterium that is motile, indole and urease negative. It is a common organism in the soil, water and can also be found on plants and animals.²⁷ The presence of *Pseudomonas aeruginosa* a predominant soil bacterium suggests transmission from settling dust suspensions.²⁸ As opined by,⁶ the presence of *Pseudomonas aeruginosa* in food stuff items if either not cooked or not properly cooked before consumption, may lead to gastroenteritis as this bacterium is often associated with gastrointestinal infection. It is therefore, very imperative that this microorganism be eliminated from food samples, and material used in food preparation to prevent bacterial infections.

The result of the antibiotic sensitivity pattern of the isolated bacteria species by disc diffusion method as shown in Table 5, indicates that Ciprofloxacin was the most effective antibiotics against the tested bacterial isolates followed by Levofloxacin, Gentamicin and Ofloxacin while Augmentin was the least. Ciprofloxacin, Levofloxacin and Ofloxacin are effective against both gram positive and negative bacteria and this correspond with the report of,¹⁰ who worked on bacterial contamination of toilet door handles on Baze University campus Abuja Nigeria. Ciprofloxacin, Levofloxacin and Ofloxacin belong to the class of antibiotics called Quinolones and are active against type II topoisomerases and act by blocking DNA replication and inhibiting synthesis and cell division.²⁹ The mechanism of quinolone inhibition occurs through formation of a ternary cleavage complex with the topoisomerase enzyme and DNA.³⁰ Gentamicin in this present research was effective against both gram positive and negative bacteria and it works by killing or preventing bacteria growth. It belongs to the class of antibiotics called Aminoglycoside. In this study, gentamicin was effective against all the tested bacterial isolates which is in accordance with the earlier reports of.^{31, 32}

Table 5: Antibiotics Sensitivity Pattern of the Isolated Bacteria species

Antibiotics	Bacterial Isolates						
	<i>S. aureus</i> (N= 24) n(%)	<i>Bacillus species</i> (N=13) n(%)	<i>E. coli</i> (N = 9) n(%)	<i>E. aerogenes</i> (N= 6) n(%)	<i>P. aeruginosa</i> (N= 4) n(%)	<i>Salmonella typhi</i> (N= 3) n(%)	<i>Streptococcus species</i> (N= 3) n(%)
CIP	24(100)	10(76.9)	9(100)	6(100)	2(50)	3(100)	2(66.7)
AUG	10(41.7)	0(0)	9(100)	0(0)	0(0)	2(66.7)	0(0)
GEN	16(66.7)	8(61.5)	7(77.8)	5(83.3)	2(50)	2(66.7)	2(66.7)
LEV	17(70.8)	11(84.6)	8(88.9)	6(100)	3(75)	3(100)	3(100)
STR	24(100)	0(0)	7(77.8)	0(0)	3(75)	3(100)	2(66.7)
OFL	19(79.2)	7(53.8)	9(100)	3(50)	2(50)	3(100)	2(66.7)
IMI	24(100)	9(69.2)	8(88.9)	1(16.7)	4(100)	3(100)	3(100)
AMO	17(70.8)	0(0)	2(22.2)	4(66.7)	0(0)	1(33.3)	3(100)
SEP	20(83.3)	10(76.9)	4(44.4)	3(50)	3(75)	3(100)	1(33.3)

CIP = Ciprofloxacin (5µg), AUG = Augmentin (30µg), GEN = Gentamicin (10µg), LEV = Levofloxacin (5µg), STR = Streptomycin (30µg), OFL = Ofloxacin (5µg), IMI = Imipenem (5µg), AMO = Amoxicillin (30µg), SEP = Septrin (30µg).

Augmentin and Amoxicillin was the least effective among the different antibiotics tested against the bacterial isolates and *Bacillus species*, *Enterobacter aerogenes*, *Streptococcus species* and *Pseudomonas*

aeruginosa was resistant (0%) each but *Escherichia coli* was 9(100%) susceptible to Augmentin. The resistance of the bacterial isolates to Augmentin in this present study is of public health concern and⁶, reported similar result, even as it is often used as a drug of choice in the treatment of bacterial infections. However, in place of it, Ciprofloxacin

and Levofloxacin may be used for the treatment of such infections, but what is of massive worry for now is the situation where the use of Augmentin and Amoxicillin which are broad spectrum antibiotics has been abused in the local communities with a resistance feedback.⁶

Conclusion

The results of this study indicated that the food items grinded with the hand manual grinders were contaminated with two or more different pathogenic bacteria which can result to spread of infectious diseases. A total of 62 bacterial species was obtained which accounted for 40 gram positive and 22 gram negative bacteria. Seven (7) bacteria species were isolated which includes *Staphylococcus aureus*, *Bacillus* species, *Escherichia coli*, *Enterobacter aerogenes*, *Pseudomonas aeruginosa*, *Salmonella typhi* and *Streptococcus* species. The susceptibility testing of the bacteria isolates against different antibiotics indicates that Ciprofloxacin, Levofloxacin, Gentamicin and Ofloxacin are the most effective against the bacteria tested while the least was Augmentin and Amoxicillin. The resistance of the bacterial isolates to Augmentin and Amoxicillin in this present study is of public health concern, as such hand manual grinders used in the market for grinding food stuffs should be properly cleaned before and after use, and individual household should have their own personal hand manual grinder, market sanitation and personal hygiene should also be observed and encouraged regularly to reduce spread of infectious diseases.

Conflict of Interest

The authors declare no conflict of interest.

Author's Declaration

The authors hereby declare that the work presented in this article is original. Any liability for claims relating to this article will be borne by us.

Acknowledgement

The study was supported by the Department of Microbiology, Delta State University of Science and Technology, Ozoro.

References

- Mba SA, Obikwelu MC, Obienusi E. Assessment of the Basic Sanitation Facilities in the Major Markets of Anambra State, Nigeria. *Socialsci. J. Soc. Sci. and Hum.* 2022; 7(2):89-97.
- Federal Ministry of Environment (FMOE). Policy Guidelines on Market and Abattoir Sanitation, Abuja, Nigeria. 2005
- WHO. UNICEF, Nigeria – evaluation report on WASH programme 2014-2017. 2017.
- Ibanga EE. An Assessment of Environmental Sanitation in an Urban Community in Southern Nigeria. *Afri. J. Envi. Sci. and Technol.* 2015; 9(7):592-599.
- Orogu JO, Ehiwario NJ, Adebisi OO. Microbiological Assessment of Cutleries. *MOJ Bioequiv. Availab.* 2017; 3(6):1-5.
- Azuonwu O, Azuonwu TC, Ndah, MA. Isolation and identification of potential high risk pathogens from blenders used in grinding some food stuffs in a local community market in rivers state: a public health concern. *J. Microbiol. & Experi.* 2019; 7(4):183–187.
- Azuonwu O, Nnenna I, Douglass AS, Ntaa BN. Consequences of haemolytic disease of the fetus and newborn (HDFN) and the clinical significance of antibody screening in prenatal diagnosis: A study of multigravida and primigravida women in Port Harcourt, Niger Delta. *J. Clin. Lab. Med.* 2016; 1(1):1–7.
- Odigie AB, Ekhiase FO, Orjiakor PI, Omozuwa S. The role of door handles in the spread of microorganisms of public health consequences in University of Benin teaching hospital (UBTH), Benin City, Edo State. *Pharm. Sci. Tech.* 2017; 2(2):15-21.
- Iskandar S, Amir S, Tarek N. Isolation of Potentially Pathogenic Bacteria from Public Service Cars Door Handles. *Int. J. Curr. Microbiol. App. Sci.* 2018; 7(12):1154-1159.
- Alonge OO, Auwal BM, Aboh MI. Bacterial Contamination of toilet door handles on Baze University campus, Abuja Nigeria. *Afri. J. Clin. and Experi. Microbiol.* 2019; 20 (1):35-41
- Baker KA, Han IY, Bailey J, Johnson L, Jones E, Knight A, MacNaughton M, Marvin P, Nolan K, Martinez-Dawson R, Dawson PL. Bacterial transfer from hands while eating popcorn. *Food Nutritn. Sci.* 2015; 6:1333-1338.
- Ventola CL. The Antibiotic Resistance Crisis: Part 1: Causes and Threats. *Pharm. and Therapeutics*, 2015; 40(4):277-283.
- Mirela V, Carmen C, Cristina EZ, Adrian V, Valentina B, Liana S, Maria S, Maria P, Veronica B. Comparative study of antimicrobials use and the antibiotic resistance of gram negative strains. *Farmac.* 2017; 65(2):225-229.
- Cheesbrough, M. *District laboratory Practice in Tropical Countries. Part 2.* Cambridge University Press; 2006; 143–157.
- Clinical and Laboratory Standards Institute (CLSI). *Performance standards for antimicrobial susceptibility testing.* 2017.
- Ginocchio CC. Role of NCCLS in antimicrobial susceptibility testing and monitoring. *Am. J. Health-Systems Pharm.* 2002; 59(8 Suppl 3):7-11.
- Herawati C, Endayani H, Indragiri S, Kistanti L, Supriatin N, Wahyuni NT, Dani AH, Hikmawati I. Sanitary Hygiene and Behavior of Food Handlers in the Presence of *Escherichia coli* Bacteria. *J Pure Appl Microbiol.* 2023; 17(4):2098-2103.
- Lowy FD. *Staphylococcus aureus* infections. *N Engl J Med.* 1998; 339(8):520-532.
- Chambers HF. Community-associated MRSA resistance and virulence converge. *N Engl J Med.* 2005; 352(14):1485-1487.
- Boucher HW, Corey GR. Epidemiology of methicillin-resistant *Staphylococcus aureus*. *Clin Infect. Dis.* 2008; 46(5):S344-349.
- Rasigade JP, Vandenesch F. *Staphylococcus aureus*: A pathogen with still unresolved issues. *Infect. Genet. Evol.* 2014; 21:510-514.
- Tong SY, Davis JS, Eichenberger E, Holland TL, Fowler VG. *Staphylococcus aureus* infections: Epidemiology, pathophysiology, clinical manifestations, and management. *Clin Microbiol Rev.* 2015; 28(3):603-661.
- Ekundayo FO, Oladipupo OA, Ekundayo EA. Studies on the effects of microbial fermentation on bush mango (*Irvingia gabonensis*) seed cotyledons. *Afri. J. Microbiol. Res.* 2013; 7(34):4363– 4367.
- Hassanien AA, Abdel-Aziz NM. Prevalence of antimicrobial-resistant *Streptococcus* species among respiratory patients and meat products, and antibacterial effects of oregano oil nanoemulsion, *Int. J. One Health*, 2021; 7(1):135-141.
- Beilharz K, Nováková L, Fadda D, Branny P, Massidda O, Veening J. Control of cell division in *Streptococcus pneumoniae* by the conserved Ser/Thr protein kinase StkP. *Proc. Natl. Acad. Sci. U. S. A.* 2012; 109(15):E905-E913.
- Enabulele O, Awunor SN. Typhoid fever in a Tertiary Hospital in Nigeria: Another look at the Widal agglutination test as a preferred option for diagnosis. *J. Nig. Med. Ass.* 2016; 57(3):145-149.
- Nicholas MM, Maribeth LF. Introduction: *Pseudomonas aeruginosa*. *Clin Lab Sci.* 2011; 24(1):41-42.
- Atiku S, Ogbaga CC, Alonge OO, Nwagbara OF. Comparative study on the physicochemical and bacteriological qualities of some drinking water sources in

- Abuja, Nigeria. *Global J. Pure and Appl. Sci.* 2018; 24(1):91-98.
29. Vila J. Fluoroquinolone resistance. In: White DG, Alekshun MN, McDermott PE (editors). ASM press 2005:41-52.
30. Hiasa H, Shea ME. DNA gyrase mediated wrapping of the DNA strand is required for the replication fork arrest by the DNA gyrase-quinolone-DNA ternary complex. *J Biol Chem.* 2000; 275: 34780-34786.
31. Appeh OG, Egwuatu TF, Nwankwo OE, Ibe AC. Bacterial Contamination of Microphones used in places of worship in Umuahia, Abia State, Nigeria. *SuanSun. Sci. Techno. J.* 2019; 6(2):16-23.
32. Amare A, Worku T, Ashagirie B, Adugna M, Getaneh A, Dagnaw M. Bacteriological profile, antimicrobial susceptibility patterns of the isolates among street vended foods and hygienic practice of vendors in Gondar town, Northwest Ethiopia: a cross sectional study. *BMC Microbiol.* 2019; 19(120):1-9.