

PREVALENCE OF *Salmonella* AND *Staphylococcus* SPECIES IN READY-TO-EAT FOOD AND DRINKING WATER SOLD WITHIN BENSON IDAHOSA UNIVERSITY LEGACY CAMPUS OHKA, BENIN CITY, EDO STATE NIGERIA

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ABSTRACT

The aim of this study was to evaluate the prevalence of *Salmonella* and *Staphylococcus* species in ready-to-eat food and drinking water sold within Benson Idahosa University Legacy Campus, Okha, Benin City, Edo State, Nigeria. All samples were obtained aseptically in a biohazard bag and transported in ice pack to the laboratory for immediate analysis. All samples were processed and enumeration of total heterotrophic bacterial count, total *Staphylococcus aureus* counts and total *Salmonella* counts were determined by pour plate method using nutrient agar, mannitol salt agar and *Salmonella-Shigella* agar respectively by standard microbiological techniques. All bacterial isolates were identified by their cultural, morphological and biochemical characteristics. Their antibiotics susceptibility profile, multidrug resistant profile and phenotypic identification of methicillin resistant *staphylococcus aureus* were determined according to the description of Clinical and Laboratory Standard Institute (CLSI). The highest bacterial count for the drinking water samples ranged from $200 \pm 45.83 \times 10^5$ cfu/ml to $126.67 \pm 25.17 \times 10^5$ cfu/ml while no *Staphylococcus aureus* and *Salmonella* was reported in all the drinking water samples collected from within the university campus. For the ready-to-eat food samples, the mean heterotrophic bacterial count obtained varied between $143.33 \pm 49.09 \times 10^5$ cfu/g to $174.67 \pm 40.46 \times 10^5$ cfu/g. Fifty (50) bacterial isolates were presumptively identified as *Salmonella* and *Staphylococcus aureus* spp. and they were resistant to ceftazidime, cefuroxime, cefixime and augmentin. The multidrug resistant index recorded was between 50 % and 75 %. Out of the 22 isolates of *Staphylococcus aureus* in ready-to-eat food samples, 21 exhibited high levels of antibiotics multiple drug resistance while 13 out of 28 *Salmonella* isolates exhibited high levels of antibiotics multiple drug resistance. Phenotypic identification revealed 21 *Staphylococcus aureus* isolates to be methicillin resistant. This study demonstrated that ready to eat food sold within Benson Idahosa University Legacy campus, Okha, are not safe for human consumption. This therefore, calls for serious attention to avoid the outbreak and spread of antibiotic resistant determinants.

KEYWORDS: Food safety; drinking water quality; antibiotics resistance; *Salmonella*; *Staphylococcus aureus*

INTRODUCTION

Ready-to-eat foods sold globally remain a source of concern, especially with respect to their public health significance (Wogu *et al.*, 2011; O'Brien, 2014; Whitworth, 2019; Lee and Yoon, 2021). This is particularly worse in developing countries because of poor knowledge of food safety practices (Chukwuocha *et al.*, 2009; Adebukola *et al.*, 2015). Amisah and Owusu (2012) reported that only 23.8% of 21 food vendors interviewed within a university campus in Ghana have had any form of formal training in food hygiene and safety practices, while less than 40% of them had no knowledge of microorganisms or food contamination. In Ogun State, South Western Nigeria, Adebukola *et al.* (2015) reported 41.6% (n = 473) of respondents to have poor knowledge of food borne infection, while only 7.6% had adequate knowledge of food borne infection. Meanwhile, in South Eastern Nigeria, 48.4% of respondents had poor knowledge of food sanitation (Chukwuocha *et al.*, 2009).

The existing gaps in public awareness of food borne diseases/outbreaks between developed and developing countries is evident from the number of food borne related disease outbreaks reported in developed countries (O'Brien, 2014; Whitworth, 2019; Lee and Yoon, 2021). In 2018 for example, EU Member States reported 5,146 foodborne outbreaks affecting 48,365 people (European Centre for Disease Prevention and Control, 2019). Such data are however scarce in most developing countries, despite existing reports on poor microbiological quality of RTE foods. In Nigeria, studies on the

microbiological safety of ready-to-eat foods from tertiary institutions are reported to be poor and of public health concern (Odu and Assor, 2013; Oranusi *et al.*, 2013; Akindele and Ibrahim, 2016).

Similarly, sachet-packed as well as bottled water are also delineated sources of human exposure to pathogenic microorganisms. Reports have implicated these water sources in the transmission of pathogenic microorganisms (Mohammed *et al.*, 2012; Epundu *et al.*, 2017; Iliyasu *et al.*, 2017). Very recently, Lawson *et al.* (2020) reported several bacterial pathogens, including *Vibrio cholerae* (etiologic agent of cholera) from table water sold within the campuses of the University of Port Harcourt, Nigeria. Putting all of these together, and from the complaints of students, the current study was therefore aimed at investigating the bacteriological quality of ready-to-eat foods and drinking water sold within the premises of the new campus of Benson Idahosa University, located in Okha Community, Benin City, Edo State, Nigeria.

MATERIALS AND METHODS

Sample Collection

A total of 15 samples (including 6 Ready-to-eat foods and 9 table water) were collected from Benson Idahosa University Legacy campus, Ohka, Benin City, Edo State, Nigeria and used for this study. The samples were collected in a bio-hazard bag and transported to the laboratory in ice-pack for immediate analysis.

Microbiological Analysis

One milliliter (1 ml) of each water samples were dispensed into 9 ml of sterile distilled water to make up ten-

fold serial dilution, from which subsequent dilutions were made. For the food samples, 25 g of each food samples were homogenized into 225 ml of sterile distilled water. One milliliter of the diluents was then used to make several other ten-fold dilutions. One milliliter (1ml) aliquot of the appropriate dilution was thereafter inoculated onto the appropriate medium by pour plate technique. The plates were incubated at 37°C for 18-24 hrs. All plates were done in triplicate.

Total Heterotrophic Count

One millilitre (1ml) of the appropriate dilution ten-fold serial dilution of all sample were done by adding 1ml of the stock sample into 9ml of distilled water in a sterile test tubes. Aliquot amount (1ml) of the appropriate dilutions (first [10^{-1}], third [10^{-3}] and fifth [10^{-5}]) were cultured on Nutrient agar (HiMedia Laboratory Pvt. Ltd. Mumbai, India) plates by pour plate method, and in triplicate. The cultured plates were incubated at 37°C for 24hours. The total number of colonies was counted from each plate using a colony counter (Labtech, England).

Total Staphylococcal Counts

One millilitre (1ml) of each sample was collected using a syringe and diluted into 9ml of sterile distilled water in a test tube. Appropriate dilutions (first [10^{-1}], third [10^{-3}] and fifth [10^{-5}]) were made, and 1ml each of the dilutions were cultured on Mannitol salt agar (Lab M Limited, United Kingdom) plates in triplicates. The plates were then incubated at 37°C for 24hours. The total number of colonies was counted using a colony counter (Labtech, England).

Isolation of Staphylococcus aureus on Mannitol Salt Agar

One millilitre (1ml) of the appropriate dilutions' 10^{-1} , 10^{-3} and 10^{-5} were inoculated into already prepared mannitol salt agar (Lab M Limited, Topley House, United Kingdom) plates using the pour plate method in triplicate. All samples were incubated at 37°C and examined after 24-48 hours for growth and change in the color of the medium. After incubation at 37°C, colonies characteristic of golden yellow, spherical, raised and smooth appearing in clusters were randomly selected and sub-cultured to obtain pure cultures for further identification and purification. The total number of colonies was counted using a colony counter (Labtech, England).

Total Salmonella Count

One millimeter (1ml) of each sample was collected using a syringe and diluted into 9ml of sterile distilled water in a test tube. Appropriate dilutions (first [10^{-1}], third [10^{-3}] and fifth [10^{-5}]) were made, and 1ml each of the dilutions were cultured on *Salmonella Shigella* agar (Titan Biotech Ltd, Rajasthan, India) plates in triplicates. The plates were then incubated at 37°C for 24hours. The total number of colonies was counted using a colony counter (Labtech, England).

Isolation of Salmonella in Salmonella Shigella Agar

Aliquot amount (1ml) of the appropriate dilutions' 10^{-1} , 10^{-3} and 10^{-5} were inoculated into already prepared *Salmonella Shigella* agar (Titan Biotech Ltd, Rajasthan, India) plates using the pour plate method in triplicate. All samples were incubated at 37°C and examined after 24-48 hours for growth

in the medium. After incubation at 37°C, colonies that appeared colorless with black centers, spherical, raised and smooth appearing in clusters were randomly selected and sub-cultured to obtain pure cultures for further identification and purification. The total number of colonies was counted using a colony counter (Labtech, England).

Presumptive Identification of Bacterial Isolates

Identification of isolates was carried out based on their morphological and biochemical characteristics.

Phenotypic identification of Methicillin-Resistant *Staphylococcus aureus* (MRSA) and Methicillin-Sensitive *Staphylococcus aureus* (MSSA)

Organisms presumptively identified as *Staphylococcus aureus* from the biochemical test were further screened for their resistance and/or sensitivity to the antibiotics methicillin as described by Fooladi *et al.* (2015) as well as Omoruyi and Ajayi (2021). Briefly, mannitol salt agar (1L) was supplemented with 0.4g of oxacillin and the isolates introduced to the medium

before being incubated at 37°C for 24hrs. Growth of *Staphylococcus aureus* on the medium (Mannitol-oxacillin medium) was indicative of Methicillin-resistance while inhibition of *Staphylococcus aureus* growth indicated Methicillin-sensitive strains of *Staphylococcus aureus*.

Statistical Analysis

All statistical analysis was performed using the SPSS software package version 23. All experiments were performed in triplicates and the results expressed as the means ± standard deviation (SD).

RESULTS

The results of the current study shows that heterotrophic bacteria, *Staphylococcus aureus* and *Salmonella* species are prevalent in ready-to-eat food and drinking water samples sold within the premises of Benson Idahosa University Legacy campus. The heterotrophic bacterial counts, *Staphylococcus aureus* counts and *Salmonella* counts are presented in Table 1.

Table 1: Total Heterotrophic, *Staphylococcus aureus* and *Salmonella* counts obtained from drinking water

Sample Code	THC (cfu/ml) (×10 ⁵)	TSAC (cfu/ml) (×10 ⁵)	TSC (cfu/ml) (×10 ⁵)
AOSW	142.33 ± 22.19	0.00 ± 0.00	0.00 ± 0.00
AOBW	193.00 ± 8.89	0.00 ± 0.00	0.0 ± 0.00
OSW	146.00 ± 39.95	0.00 ± 0.00	0.00 ± 0.00
SSW	139.33 ± 29.00	0.00 ± 0.00	0.00 ± 0.00
FSW	170.00 ± 12.53	0.00 ± 0.00	0.00 ± 0.00
SBW	158.33 ± 34.03	0.00 ± 0.00	0.00 ± 0.00
OLSW	164.00 ± 22.61	0.00 ± 0.00	0.00 ± 0.00
NDSW	200.00 ± 45.83	0.00 ± 0.00	0.00 ± 0.00
EBW	126.67 ± 25.17	0.00 ± 0.00	0.00 ± 0.00

KEY: THC: Total Heterotrophic count; TSA: Total *Staphylococcus aureus* count; TSC: Total *Salmonella* count

The highest bacterial count for the drinking water samples was observed in NDSW ($200 \pm 45.83 \times 10^5$ cfu/ml); while the lowest heterotrophic count was observed in EBW ($126.67 \pm 25.17 \times 10^5$ cfu/ml). Interestingly, no *Staphylococcus aureus* and *Salmonella* were reported in all the drinking water samples. The mean heterotrophic bacterial count obtained from all food samples ranged between $143.33 \pm 49.09 \times 10^5$ cfu/g to $174.67 \pm 40.46 \times 10^5$ cfu/g as shown in Table 2.

The total *Staphylococcus aureus* count were fairly low in all the food

samples investigated, with mean *Staphylococcus aureus* counts ranging between 0.00 to $73.33 \pm 7.64 \times 10^5$ cfu/g. Meanwhile, no *Staphylococcus aureus* was reported in S03, B04, R05 and FF06 samples investigated. The total *Salmonella* counts (TSCs) obtained were a little higher in FB01 ($300 \pm 0.00 \times 10^5$ cfu/g), when compared with the TSCs obtained from BE02 (62.33 ± 26.73 cfu/g) [Table 2]. Also, as with *Staphylococcus aureus*, no *Salmonella* was reported in S03, B04, R05 and FF06 samples collected from within the university campus.

Table 2: Total heterotrophic, *Staphylococcus aureus* and *Salmonella* counts obtained from ready-to-eat food samples

Sample Code	THC (cfu) ($\times 10^5$)	TSAC (cfu) ($\times 10^5$)	TSC (cfu) ($\times 10^5$)
FB01	161.00 ± 11.53	73.33 ± 7.64	300.00 ± 0.00
BE02	161.33 ± 53.27	75.67 ± 17.16	62.33 ± 26.73
S03	170.00 ± 36.06	0.00 ± 0.00	0.00 ± 0.00
B04	174.67 ± 40.46	0.00 ± 0.00	0.00 ± 0.00
R05	163.00 ± 33.45	0.00 ± 0.00	0.00 ± 0.00
FF06	143.33 ± 49.09	0.00 ± 0.00	0.00 ± 0.00

KEY: THC: Total Heterotrophic count; TSA: Total *Staphylococcus aureus* count; TSC: Total *Salmonella* count; FB: Fried beef; BE: Boiled egg; S: Salad; B: Beans; R: Rice; FF: Fried fish

Table 3: Cultural, morphological and biochemical characteristics of the isolates

Characteristics	Presumptive Identity	
	<i>Salmonella</i>	<i>Staphylococcus aureus</i>
Gram Reaction	-ve	+ve
Catalase	+ve	+ve
Oxidase	-ve	-ve
Motility	+ve	-ve
Citrate	+ve	+ve
Indole	-ve	-ve
Total number of Isolates	28	22

KEY: +ve; Positive, -ve: Negative

Table 3 shows the morphological and biochemical characteristics of the presumptive isolates, which included *Salmonella* and *Staphylococcus aureus*. Based on their presumptive identity, 28

isolates were observed to be *Salmonella* species while 22 isolates were observed to be *Staphylococcus aureus* (Table 3).

The selected isolates (*Staphylococcus aureus* and *Salmonella*

species) were tested for their sensitivity and/or resistance to the following antibiotics: gentamicin (10µg), nitrofurantion (300µg), augmentin (30µg), ofloxacin (5µg), cefixime (5µg), ciprofloxacin (5µg), ceftazidime (30µg), cefuroxime (30µg) and oxacillin (1µg). All *Staphylococcus aureus* isolates were observed to be resistant to ceftazidime, cefuroxime, cefixime and augmentin (Table 4). Phenotypic identification of methicillin resistant *Staphylococcus aureus* further showed 21 of the 22 isolates to be methicillin resistant (Table 4).

The antibiotics susceptibility profile also shows all *Salmonella* isolates to be resistant to ceftazidime, cefuroxime, cefixime and augmentin (Table 5). Their multidrug resistant index for *Staphylococcus aureus* and *Salmonella* species ranged between 50% and 75% as reflected in Tables 7 and Table 8. nBE13 had the highest AMDR profile while BE04 had the least AMDR profile for *Staphylococcus aureus* (Table 6), while *Salmonella* species isolated from BE03; BE07; BE05 and BE12 had the highest AMDR profile (Table 7).

Table 4: Antibiotics susceptibility pattern of *Staphylococcus aureus* from ready-to-eat food samples

ISOLATE CODE	ANTIBIOTICS									MRSA/MSSA	MDR INDEX (%)
	CAZ	CRX	GEN	CXM	OFL	AUG	NIT	CPR	OXA		
FB01	R	R	S	R	S	R	S	S	R	MRSA	50
FB02	R	R	S	R	S	R	S	S	R	MRSA	50
FB03	R	R	S	R	I	R	S	S	R	MRSA	50
FB04	R	R	I	R	S	R	I	S	R	MRSA	50
FB05	R	R	S	R	S	R	I	S	R	MRSA	50
FB06	R	R	S	R	I	R	I	S	R	MRSA	50
FB07	R	R	I	R	I	R	S	S	R	MRSA	50
BE01	R	R	I	R	S	R	S	S	R	MRSA	50
BE02	R	R	S	R	S	R	S	S	R	MRSA	50
BE03	R	R	R	R	S	R	S	S	R	MRSA	62.5
BE04	R	R	I	R	S	R	S	S	I	ND	50
BE05	R	R	I	R	S	R	S	I	R	MRSA	50
BE06	R	R	I	R	I	R	S	I	R	MRSA	50
BE07	R	R	I	R	S	R	S	I	R	MRSA	50
BE08	R	R	S	R	S	R	S	I	R	MRSA	50
BE09	R	R	I	R	S	R	R	I	R	MRSA	62.5
BE10	R	R	S	R	S	R	R	I	R	MRSA	62.5
BE11	R	R	S	R	S	R	R	I	R	MRSA	62.5
BE12	R	R	S	R	S	R	S	I	R	MRSA	50
BE13	R	R	R	R	S	R	R	S	R	MRSA	75
BE14	R	R	S	R	S	R	S	R	R	MRSA	62.5
BE15	R	R	I	R	S	R	S	R	R	MRSA	62.5

KEY: CAZ: Ceftazidime CRX: Cefuroxime, GEN: Gentamicin, CXM: Cefixime, OFL: Ofloxacin, AUG: Augmentin, NIT: Nitrofurantion, CPR: Ciprofloxacin, OXA: Oxacillin, R- Resistance; S- Sensitive; I- Intermediate; FB: Fried beef; BE: Boiled egg; MRSA: Methicillin-resistant *Staphylococcus aureus*, MDR: Multi drug resistance index.

Table 5: Antibiotics susceptibility pattern of *Salmonella* from ready-to-eat food samples

ISOLATE CODE	ANTIBIOTICS								MDR INDEX (%)	
	CAZ	CRX	GEN	CXM	OFL	AUG	NIT	CPR		
FB01		R	R	S	R	S	R	S	S	50
FB02		R	R	S	R	I	R	I	S	50
FB03		R	R	I	R	R	R	I	I	62.5
FB04		R	R	I	R	S	R	S	S	50
FB05		R	R	I	R	S	R	S	S	50
FB06		R	R	S	R	S	R	S	S	50
FB07		R	R	R	R	S	R	S	S	62.5
FB08		R	R	R	R	S	R	S	S	62.5
FB09		R	R	R	R	S	R	S	S	62.5
FB10		R	R	R	R	S	R	S	S	62.5
FB11		R	R	R	R	S	R	S	I	62.5
FB12		R	R	I	R	S	R	S	S	50
FB13		R	R	S	R	I	R	S	S	50
FB14		R	R	I	R	I	R	S	S	50
FB15		R	R	I	R	I	R	S	S	50
BE01		R	R	I	R	S	R	I	S	50
BE02		R	R	I	R	S	R	S	R	62.5
BE03		R	R	S	R	S	R	R	R	75
BE04		R	R	I	R	S	R	S	I	50
BE05		R	R	R	R	I	R	I	R	75
BE06		R	R	R	R	I	R	I	S	62.5
BE07		R	R	I	R	S	R	R	R	75
BE08		R	R	S	R	S	R	S	R	62.5
BE09		R	R	S	R	S	R	S	I	50
BE10		R	R	S	R	S	R	S	S	50
BE11		R	R	S	R	S	R	I	S	50
BE12		R	R	R	R	S	R	R	S	75
BE13		R	R	S	R	I	R	S	S	50

KEY: CAZ: Ceftazidime CRX: Cefuroxime, GEN: Gentamicin, CXM: Cefixime, OFL: Ofloxacin, AUG: Augmentin, NIT: Nitrofurantion, CPR: Ciprofloxacin, R- Resistance; S- Sensitive; I- Intermediate; FB: Fried beef; BE: Boiled egg; MDR: Multi drug resistance index

Table 6: AMDR profile of *Staphylococcus aureus* isolated from ready-to-eat

NO OF ISOLATES	AMDR PROFILE							ISOLATE CODE
1	CAZ	CRX	CXM	AUG				BE04
7	CAZ	CRX	CXM	AUG	OXA			FB01; FB02; FB03; FB04; FB05; FB06; FB07
7	CAZ	CRX	CXM	AUG	OXA			BE01; BE02; BE05; BE06; BE07; BE08; BE12
1	CAZ	CRX	GEN	CXM	AUG	OXA		BE03
3	CAZ	CRX	CXM	AUG	NIT	OXA		BE09; BE10; BE11
2	CAZ	CRX	CXM	AUG	CPR	OXA		BE14; BE15
1	CAZ	CRX	GEN	CXM	AUG	NIT	OXA	BE13

KEY: CAZ: Ceftazidime CRX: Cefuroxime, GEN: Gentamicin, CXM: Cefixime, OFL: Ofloxacin, AUG: Augmentin, NIT: Nitrofurantion, CPR: Ciprofloxacin, OXA: Oxacillin FB: Fried beef; BE: Boiled egg; AMDR: Antibiotics Multiple drug resistance profile

Table 7: AMDR profile of *Salmonella* isolated from ready-to-eat food samples

NO OF ISOLATES	AMDR PROFILE							ISOLATE CODE
9	CAZ	CRX	CXM	AUG				FB01; FB02; FB04; FB05; FB06; FB12; FB13; FB14; FB15
6	CAZ	CRX	CXM	AUG				BE01; BE04; BE09; BE10; BE11; BE13
1	CAZ	CRX	CXM	OFL	AUG			FB03
6	CAZ	CRX	GEN	CXM	AUG			FB07; FB08; FB09; FB10; FB11; BE06
2	CAZ	CRX	CXM	AUG	CPR			BE02; BE08
1	CAZ	CRX	CXM	AUG	NIT	CPR		BE03; BE07
1	CAZ	CRX	GEN	CXM	AUG	CPR		BE05
1	CAZ	CRX	GEN	CXM	AUG	NIT		BE12

KEY: CAZ: Ceftazidime CRX: Cefuroxime, GEN: Gentamicin, CXM: Cefixime, OFL: Ofloxacin, AUG: Augmentin, NIT: Nitrofurantion, CPR: Ciprofloxacin, FB: Fried beef; BE: Boiled egg; AMDR: Antibiotics Multiple drug resistance profile

DISCUSSION

Drinking water sources and ready-to-eat foods sold on campuses are poorly delineated sources of human exposure to pathogenic microorganisms. In Benson Idahosa University Legacy campus, Okha Community, Edo State, South-South Nigeria, there have been complaints by patrons and students on the poor quality of RTE foods served in such facilities, and the potential of the food to contain food borne pathogen. To further substantiate their claim, we investigated the prevalence of two potential foodborne pathogens (*Staphylococcus aureus* and *Salmonella* species) in RTE foods and drinking water samples sold within the School premises.

The result shows that heterotrophic bacteria are prevalent in both sachet-packed and bottled water sold within Benson Idahosa University Legacy campus. However, the two pathogens (*Staphylococcus aureus* and *Salmonella* species) of interest were absent in all water samples investigated. Their absence in drinking water samples is an indication of good manufacturing practices. This is in keeping with the results of Hillo (2016), who reported the absence of *S. aureus* in drinking water samples sold in Basrah, Iraq.

Elsewhere, Lawson *et al.* (2020) reported *Staphylococcus aureus* and other pathogenic Gram negative bacteria such as *Escherichia* species and *Vibrio cholerae* from table water samples sold on School campuses of the University of Port Harcourt, Nigeria. Results of the microbiological assessment of sachet-packed and bottled water across different cities in Nigeria shows that approximately 50% of the samples are unfit for human consumption (Ibrahim *et al.*, 2015). This is however worrisome, considering the high dependence on sachet-packed and bottled water in Nigeria and other developing countries. This dependence is attributed to government failure to provide safe and potable drinking water for her populace (Lawson *et al.*, 2020).

Meanwhile, only two RTE foods (fried beef and boiled egg) were observed to have the potential food borne pathogens (*Staphylococcus aureus* and *Salmonella* spp) as well as heterotrophic bacteria. Their presence may have occurred inadvertently during the processing of the food items. Ready-to-eat food samples sold within University campuses in Nigeria have previously been reported to be contaminated with heterotrophic bacteria and other pathogens (Table 8).

Table 8: Microbiological Quality of Ready-to-Eat Foods sold within different University Campuses in Nigeria

Food	Microbial count	Microbial pathogen (from all samples)	University	State	Geo-Political Zone	Reference
Rice	1.0 – 51 x 10 ³	<i>Escherichia coli</i> , <i>Bacillus cereus</i> , <i>Salmonella</i> spp	Ekiti State University	Ekiti	South West	Akindele and Ibrahim, 2016
Beans	2.4 – 60 x 10 ²	<i>Clostridium perfringens</i> , <i>Shigella</i> spp				
Yam	1.3 – 36 x 10 ²	<i>Klebsiella</i> spp., <i>Proteus</i> spp				
“Fufu”	3.1 – 55 x 10 ³	<i>Staphylococcus aureus</i> , <i>Campylobacter</i> spp				
Meat	NG – 6.0 x 10 ⁴	<i>Aspergillus</i> spp., <i>Mucor</i> spp. <i>Rhizopus</i> spp				
“Coleslaw”	ND	<i>Bacillus</i> spp., <i>E. coli</i> ,	Federal University of Technology, Owerri	Imo	South East	Oranusi <i>et al.</i> , 2013
Fried rice	ND	<i>Klebsiella</i> spp., <i>Proteus</i> spp <i>Staphylococcus aureus</i>				
Jollof rice	ND	<i>Aspergillus niger</i> , <i>Aspergillus fumigates</i>				
Moi-moi	ND	<i>Penicillium</i> spp. <i>Mucor</i> spp				
Hands of Vendors	76 – 88 cfu	<i>Klebsiella</i> spp., <i>Enterobacter</i> spp, <i>staphylococcus</i> spp. <i>Proteus</i> sp <i>E. coli</i> , <i>Salmonella</i> spp., <i>Micrococcus</i> sp.,	Rufus Giwa Polytechnic	Ondo	South West	Ibrahim <i>et al.</i> , 2013
Table	08 – 184 cfu	<i>Bacillus</i> spp., <i>Pseudomonas</i> and <i>Streptococcus</i> spp., <i>Serratia</i> spp.				
Rice	2.5 – 17.8x10 ⁵	<i>Bacillus cereus</i> , <i>S. aureus</i>	University of Port Harcourt	Rivers	South South	Odu and Assor, 2013
Beans	3.5 – 17.1x10 ⁴	<i>E. coli</i> , <i>Klebsiella Pneumoniae</i>				
Rice	5.2 – 10 x 10 ⁷	<i>S. aureus</i> , <i>Streptococcus pneumoniae</i> , <i>E.</i> <i>coli</i>	Federal University, Dutse	Jigawa	North West	Muhammad <i>et al.</i> , 2016
Beans pudding	3.2 – 8.4. x 10 ⁶	<i>Salmonella</i> spp				
Pounded yam	3.6 – 11 x 10 ⁶	<i>Bacillus cereus</i>				
Local cheese	2.8 – 9.2 x 10 ⁶					
Beans cake	4.8 – 7.2 x 10 ⁶					
Rice	3.0 – 4.5 x 10 ⁴	<i>S. aureus</i> , <i>E. coli</i>	Federal Polytechnic Bali	Taraba	North East	Monday <i>et al.</i> , 2014
Moi-moi	4.0 – 8.7 x 10 ⁴	<i>Klebsiella</i> spp., <i>Salmonella</i> spp., <i>Mucor</i> spp., <i>Aspergillus</i> spp				

Salmonellosis and staphylococcal food poisoning are two common types of food borne diseases reported to be prevalent in both developed and developing countries. The presence of *Salmonella* species in boiled eggs sold within Benson Idahosa University is also not unexpected, as this organism have previously been reported to be prevalent in boiled eggs (Moosavy *et al.*, 2015; Nagappa *et al.*, 2007). In 2018, Slovakia, Spain and Poland accounted for 67% of all *Salmonella* outbreaks in the EU, and the outbreaks were mainly linked to eggs (European Centre for Disease Prevention and Control, 2019). In Iran and Indian, the prevalence are reported to be rather low; 1.33% and 1.5% respectively (Moosavy *et al.*, 2015; Nagappa *et al.*, 2007). Globally, eggs are reported to be a major source of *Salmonella* infection and are contaminated more frequently with this organism when the egg shells are dirty and/or cracked as well as stored above 8°C (Little *et al.*, 2017). However, boiling raw eggs for 5 mins after the water starts to boil as well as cooking in the microwave oven have been reported to be completely effective in the elimination of *Salmonella* contamination from eggs (Savi *et al.*, 2011).

On the other hand, the presence and consumption of *Staphylococcus* species in RTE food may lead to staphylococcal food poisoning. The presence of this organism in some RTE food samples investigated in the current study is an indication of poor handling and processing technique. This result is in agreement with a similar study that reported *S. aureus* in RTE foods, with the source of contamination being the

hands of the food handlers (Syne *et al.*, 2013). Furthermore, studies by Claudia and Maria, (2013) also reported a similar outcome in RTE foods bought at the point of sale and raw fruits and vegetables ready for consumption. The result of Mensah *et al.* (2002) and Sina *et al.*, (2011) have also reported salad and macaroni dishes in Accra, Ghana to have high levels of *S. aureus*, which is contrary to the results obtained in the current study. In the East Asian region, studies in Korea have shown that RTE foods are regularly contaminated with *S. aureus*. Normanno *et al.* (2005) reported *S. aureus* as the most frequent contaminant in cream cakes. This may be due to the content of dairy products such as cream which are often the main cause of the occurrence of staphylococcal food poisoning. According to Colombari *et al.* (2007), RTE food, especially salads and sandwiches are among the main causes of the incident or outbreak of foodborne illness because this category of food is often prepared by hand and served cold, which may increase the incidence of contamination with potential foodborne pathogens. The high incidence of heterotrophic bacteria in RTE foods sold in Benson Idahosa University legacy campus may be due to the poor personal hygiene of most vendors and the state of the environment where these food vendors stay to carry out their activities. The role of food vendors in determining the microbial loads in food cannot be over emphasized as they contribute to the well-being of the populace with regards to their activities. In Nigeria most food vendors lack knowledge of proper food handling and may play a role in the transmission of

food borne pathogens (Adebukola *et al.*, 2015). Also, an important consideration is the fact that most food handlers do not practice good personal hygiene and good manufacturing practices because of lack of continuous awareness and education by the relevant authorities concerned with food safety regulations and also the consumers' carefree attitude to sometimes eat whatever comes their way, whether hygienically safe or not. Antibiotics resistance is a major challenge globally, but is worse in developing countries where self-medication and inappropriate use of antibiotics is a major challenge (Omoruyi *et al.*, 2020a). Clinical management of Staphylococcal infection is relied on antibiotic treatment which often fails due to aggressive resistance of organisms to antibiotics. In the current study all *Staphylococcus aureus* isolates were resistant to ceftazidime, cefuroxime, cefixime and augmentin. The bacterium was also highly sensitive to ofloxacin, this may be attributed to the fact that these antibiotics were able to penetrate the cell wall membrane and damage the nucleic acid of the isolates (Nwachukwu and Nwaigwe, 2013). This finding is similar to the results recently reported by Omoruyi *et al.*, 2020a and Omoruyi *et al.*, 2020b, who reported high prevalence of multidrug resistant *Staphylococcus aureus* from poultry and abattoir facilities respectively. Interestingly, *Salmonella* isolates also showed high multidrug resistance to ceftazidime, cefuroxime, cefixime and augmentin. The emergence of *Salmonella* with antimicrobial resistance is mainly

promoted by the use of antibiotics in animal feed to promote the growth of food animals, and in veterinary medicine to treat bacterial infections in those animals. This poses a high risk of zoonotic disease with the transmission of MDR *Salmonella* strains from animals to humans via the ingestion of food or water contaminated with the animals' faeces, direct contact or the consumption of infected food animals. Foodborne transmission of Methicillin Resistant *Staphylococcus aureus* (MRSA) is a global concern and therefore the prevalence and genetic characteristics of these organisms need to be thoroughly studied. Human infections caused by foodborne MRSA strains have been reported (Jones *et al.*, 2002). Therefore, the potential role of food in the dissemination of successful MRSA lineages cannot be ignored. In the current study all *Staphylococcus aureus* isolates tested for methicillin resistance were observed to be phenotypically positive. Song *et al.* (2015) also reported a high prevalence of MRSA in RTE food sold in china. In a contrary study, Carfora *et al.* (2015) reported low prevalence of MRSA in dairy products from Italy and in retail foods from China. Similar outcomes have also been reported in commercially processed meat products (Omoruyi and Ajayi, 2021), poultry birds (Omoruyi *et al.* 2020a) and abattoir facilities (Omoruyi *et al.*, 2020b). The presence of MRSA in RTE foods is the result of human contamination through poor personal hygiene, or through cross contamination of carcasses during food processing.

CONCLUSION

Ready-to-eat food and drinking water sold in Benson Idahosa University legacy campus Ohka, Benin city, Edo State, Nigeria analyzed were found to contain notable amount of heterotrophic bacteria per 100ml of water sample, while, no *Staphylococcus* and *Salmonella* species were detected. Meanwhile, the contamination of RTE foods by *Staphylococcus* and *Salmonella* species were common, with the majority of *Staphylococcus aureus* found to be methicillin resistant. Efforts must therefore be made to improve the hygienic conditions of food handlers to avoid the outbreak of multi-drug resistant foodborne diseases.

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