

# Screening of Kunun-zaki for Methicillin-Resistant *Staphylococcus aureus* (MRSA), Vancomycin-Resistant *Staphylococcus aureus* (VRSA) and Extended-Spectrum Beta-Lactamase (ESBL) Producing *Salmonella* spp.

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**Abstract.** Kunun-zaki is an indigenous, fermented, non-alcoholic cereal-based beverage produced and consumed primarily in Northern Nigeria. Due to its high nutritional and moisture content, it provides an ideal environment for the growth of foodborne pathogens. This study determined the presence of Methicillin resistant *Staphylococcus aureus* (MRSA), vancomycin resistant *Staphylococcus aureus* (VRSA) and Extended spectrum beta lactamase (ESBL) producing *Salmonella* spp in Kunun-zaki sold in Umuahia metropolis, Nigeria. Out of the 40 Kunun-zaki samples examined, *Salmonella* spp was detected in 19 (47.5%), whereas *S. aureus* was detected in 22 (55%) of the samples. The total *Salmonella* count ranged from  $1.90 \times 10^5$ - $9.70 \times 10^6$  (CFU/ml), whereas the total *S. aureus* count ranged from  $3.40 \times 10^4$ - $7.50 \times 10^6$  (CFU/ml); these microbial counts do not conform to the standard limits of detectable microbes in ready-to-eat food samples as stipulated by the Center for Food Safety. All the isolates were subjected to antibiotic susceptibility testing using the modified Kirby Bauer disc diffusion method. On the one hand, most of the *Salmonella* spp were resistant to the beta-lactam class of antibiotics used (IMP 58%, MEM 90%, AMP 53%). On the other hand, the *Salmonella* isolates showed the highest sensitivities to ceftazidime (95%) and ofloxacin (90%). The *S. aureus* isolates revealed high susceptibility to ofloxacin (96%), and major resistance to oxacillin (41%) and the oxyimino-cephalosporin antibiotics used (CAZ 55%, CTR 41%). MRSA, VRSA and ESBL producing *Enterobacteriaceae* are amongst the most critical multidrug-resistant bacterial pathogens. In this study, 26% of the *Salmonella* spp isolates were confirmed to be ESBL producers, whereas 41% of the isolates were MRSA, 23% were VRSA, and 68% were Vancomycin intermediate *Staphylococcus aureus* (VISA). Hence, the Kunun-zaki could constitute a reservoir for the dissemination of multidrug-resistant foodborne pathogens if not correctly quality controlled.

**Keywords:** Kunun-zaki; *Staphylococcus aureus*; *Salmonella* spp; Extended spectrum beta-lactamase (ESBL); Methicillin-resistant *Staphylococcus aureus* (MRSA); Vancomycin-resistant *Staphylococcus aureus* (VRSA); Multidrug resistance (MDR).

## INTRODUCTION

Kunun-zaki is a fermented, non-alcoholic, non-carbonated and refreshing cereal-based beverage widely consumed in Nigeria, particularly in Northern Nigeria [1]. Kunun-zaki gets its appeal from the distinctive sweet-and-sour taste characteristic of African food products fermented by Lactic Acid Bacteria (LAB) [2], as well as its purported medicinal, nutritional, and thirst-quenching properties [3, 4, 5].

Traditionally, the production procedure of Kunun-zaki varies depending on the taste and culture of the producers and consumers, thus leading to variation in the quality and stability of the Kunun-zaki. Generally, Kunun-zaki is produced from the following cereal substrates; millet, maize, wheat, rice, *acha* (*Digitaria exilis*) and sorghum, but millet is mostly used for commercial production [3, 6]. It is usually flavoured with a combination of spices such as ginger, cloves, black pepper, cinnamon, red pepper and sweeteners such as sugar and honey, together with small quantities of saccharifying agents such as paste of sweet potato tubers, malted rice and malted sorghum [6]. Kunun-zaki is relatively cheap to purchase because the ingredients and cereals used in its production are locally sourced as they are grown throughout the savannah belt of West Africa [7].

The preparation method for Kunun-zaki involves cleaning the grains of the cereals and steeping them in water for 1-3 days. This is then followed by grinding the steeped grains into a mash. The mash of cereals is then divided into two portions in a ratio of about 1:2, the larger portion is gelatinized with hot water whilst, the smaller portion is mixed with spices and the saccharifying agent. These ingredients are usually not quantified [8]. The two parts are then combined at 70-75 °C, and the mixture is left to ferment at room temperature for 18-24 hours. The product is filtered, and sweeteners are added to the filtrate to taste, and the Kunun-zaki is now ready for consumption [3, 9]. The drink is usually packaged and sold in 500ml plastic bottles (obtained from used water or carbonated drinks) and at times, even tied in some disposable polyethene bags [10].

Kunun-zaki has a concise shelf life of about 24 hours [2], readily undergoing microbial spoilage due to poor keeping conditions. Attempts to improve the shelf-life have been met with some success; pasteurization of packaging bottles followed by refrigeration storage of the Kunun-zaki

prolonged the shelf life to 8 days [11], whilst using sodium benzoate (a known preservative) followed by refrigeration extended the shelf to 21 days [12].

Studies have shown that Kunun-zaki has an essential role in the dietary pattern of its consumers owing to its richness in carbohydrates, vitamins, minerals, and moisture content. However, it is low in total protein content [4, 5]. The health benefits of Kunun-zaki includes but is not limited to: reduction of risk associated with diabetes, lowering of blood cholesterol, prevention of blood clotting [13], and increased lactation in nursing mothers [14].

Considering that Kunun-zaki is a nutritionally enriched food product with a very high moisture content and lacks standardization and quality control in both sourcing of cereals, processing, storage, handling, packaging, and delivery to customers, Kunun-zaki provides an ideal environment for the growth of foodborne microbial pathogens which include members of the *Enterobacteriaceae*, *Staphylococcus aureus*, and *Bacillus cereus*, among others, hence, predisposing consumers to serious health risks [15, 16].

*S. aureus* is a normal flora of the human skin, throat, nasal passage, fingernails, and hair. However, *S. aureus* is implicated in several infections, including meningitis, septicemia, pneumonia, endocarditis and osteomyelitis [17]. When ingested with food, *S. aureus* can cause food poisoning due to its ability to produce several enterotoxins. Also, *S. aureus* may even cause scalded skin syndrome and toxic shock syndrome through the production of different toxins [18]. *S. aureus*, in general, is sensitive to many antibiotics; however, in the last few decades, many strains of *S. aureus* have developed resistance to these antibiotics. *S. aureus* resistance to methicillin is of severe public health concern due to its high morbidity and mortality [19]. Methicillin-resistant *Staphylococcus aureus* (MRSA) occurs as a result of the acquisition of Staphylococcal Cassette Chromosome *mec* (SCC*mec*) genes by methicillin-sensitive *S. aureus*. The SCC*mec* is a mobile genetic element which carries the *mecA* or *mecC* gene which encodes the penicillin-binding protein (PBP2a) that confers resistance to all beta-lactam antibiotics [20-22]. The SCC*mec* also contains site-specific recombinase genes (cassette chromosome recombinase (*ccr*)) *ccrAB* or/and *ccrC* which mediates integration and excision of SCC*mec* into or from the chromosome. In addition, the SCC*mec* contains a few other genes and various other mobile genetic

elements such as insertion sequences, transposons, and plasmids [23, 24]. With the emergence of MRSA, vancomycin has been utilized as the treatment of choice for MRSA infections, and its excessive use has led to the emergence of Vancomycin Intermediate and Vancomycin-Resistant *S. aureus* (VISA and VRSA) [25].

Contamination of food by *Enterobacteriaceae* is usually through faecal water contamination, and cross-contamination may occur during further processing and preparation. The *Enterobacteriaceae* consists of some essential public health pathogens that produce extended-spectrum beta-lactamases (ESBL), which are a group of enzymes conferring broad resistance to beta-lactams: oxymino-cephalosporins (such as cefotaxime, ceftriaxone, ceftazidime or cefepime) and monobactams (aztreonam) but not carbapenems [26, 27]. The enzyme generally results from a point mutation in the genes of broad-spectrum beta-lactamases, TEM-1 and SHV-1, by one to four amino acid changes which form the bases of resistance [28]. ESBL enzymes are usually located on plasmids that often carry other resistance genes that confer reduced susceptibility to other unrelated classes of antimicrobials making it difficult to treat infections caused by ESBL producing bacteria [29, 30], and resistance can be easily transmitted among members of the *Enterobacteriaceae* through plasmid-mediated transfer. An essential member of the *Enterobacteriaceae* is *Salmonella* spp, which typically resides in the intestinal tract of animals and humans and are shed through faeces.

In developing countries like Nigeria, control over the processing and sales of hawked foods have not been achieved presumably because most of the vendors and sellers lack adequate knowledge of food processing and handling practices. As a result, extensive studies have been conducted on the nutritional and microbial quality of Kunun-zaki and other hawked indigenous drinks and beverages to increase public awareness, and draw the attention of the relevant food authorities to the health risks and hazards associated with the consumption of such contaminated food products [15, 31-34]. Following up from our previous study [15], in this study, we aimed to determine the presence of methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-resistant *Staphylococcus aureus* (VRSA) and extended-spectrum beta-lactamase (ESBL) producing *Salmonella* spp in Kunun-zaki drinks sold in Umuahia metropolis, Nigeria.

## MATERIALS AND METHODS

### Sample collection

Forty (40) samples of Kunun-zaki were purchased randomly from different hawkers within Umuahia metropolis, Nigeria. The samples were transported to the laboratory in an icebox within an hour of collection and were preserved in a refrigerator at 4 °C before being analyzed. The samples were analyzed within two hours of collection.

### Media used

All the media used for this study were obtained from Titan Biotech Ltd. Rajasthan, India. The media were prepared according to the manufacturers' instructions.

### *Salmonella* spp identification and characterization

An aliquot of each sample (1 ml) was enriched in a 1:10 ratio of peptone water at 37°C for 24 hours. The enriched samples were serially diluted up to 10<sup>5</sup> and 0.1ml of the appropriate dilutions were spread plated on *Salmonella Shigella* Agar (SSA) and incubated at 37°C for 24 hours. Presumptive distinct colonies (SSA: black-centred colonies) were counted and expressed as *Salmonella* spp colony forming unit per millilitre (CFU/ml) of each Kunun-zaki sample. Single colonies of *Salmonella* spp were sub-cultured for 24 hours at 37°C on nutrient agar to obtain pure cultures for biochemical identification and antimicrobial testing. Presumptive colonies were tested for the biochemical properties of *Salmonella* spp (oxidase reaction, citrate utilization test, indole production test, urease test, triple sugar iron test and H<sub>2</sub>S production).

### *Staphylococcus aureus* identification and characterization

An aliquot of each sample (1 ml) was enriched in a 1:10 ratio of peptone water at 37 °C for 24 hours. The enriched samples were serially diluted up to 10<sup>5</sup> and 0.1ml of the appropriate dilutions were spread plated on Mannitol-salt Agar (MSA) and incubated at 37°C for 24 hours. Presumptive distinct colonies (MSA: yellow colonies with yellow zones) were counted and expressed as *S. aureus* colony forming unit per millilitre (CFU/ml) of each Kunun-zaki sample. Single colonies of *S. aureus* were sub-cultured for 24 hours at 37°C on

nutrient agar to obtain pure cultures for biochemical identification and antimicrobial testing. Presumptive colonies were tested for the biochemical properties of *S. aureus* (catalase reaction, coagulase test, citrate utilization test, oxidase reaction, urease, and indole production test).

### ***Salmonella* spp antibiotic resistance profiles**

To assess the antibiotic resistance profiles of the *Salmonella* isolates, standard inoculums of the isolates were prepared from the pure cultures of *Salmonella* spp as described by Cheesbrough [35]. Standardized inoculums (one loopful) of each of the samples were subjected to susceptibility testing on Mueller Hinton Agar against 12 antimicrobial agents according to the Clinical and Laboratory Standards Institute (CLSI) protocol and criteria [36]. The following antibiotics were used: ceftazidime (CAZ; 30 µg), cefuroxime (CRX; 30 µg), gentamicin (GEN; 10 µg), ciprofloxacin (CPR; 5 µg), ofloxacin (OFL; 5 µg), augmentin (AUG; 30 µg), nitrofurantoin (NIT; 300 µg), ampicillin (AMP; 10 µg), aztreonam (ATM; 30 µg), ceftriaxone (CTR; 30 µg), imipenem (IMP; 10 µg) and meropenem (MEM; 10 µg) (Rapid labs. Ltd. Colchester, Essex, United Kingdom).

### ***Staphylococcus aureus* antibiotic resistance profiles: Methicillin-resistant *Staphylococcus aureus* and Vancomycin-resistant *Staphylococcus aureus***

To assess the antibiotic resistance profiles of the *S. aureus* isolates, standard inoculums of the isolates were prepared from the pure cultures of *S. aureus* as described by Cheesbrough [35]. Standardized inoculums (one loopful) of each of the samples were subjected to susceptibility testing on Mueller Hinton Agar against ten antimicrobial agents according to the Clinical and Laboratory Standards Institute (CLSI) protocol and criteria [36]. The following antibiotics were used: ceftazidime (CAZ; 30 µg), ceftriaxone (CTR; 30 µg), gentamicin (GEN; 10 µg), ciprofloxacin (CPR; 5 µg), ofloxacin (OFL; 5 µg), augmentin (AUG; 30 µg), erythromycin (ERY; 5 µg), vancomycin (VAN; 10 µg), oxacillin (OXA; 5 µg) and cloxacillin (CXC; 5 µg) (Rapid labs. Ltd. Colchester, Essex, United Kingdom).

### **Detection of Extended spectrum beta-lactamases (ESBL) producing *Salmonella* spp**

For detection of ESBL producing *Salmonella* spp, isolates with zones of inhibition (in diameters) of <25 mm, <22 mm and <27 mm for ceftriaxone (CTR; 30 µg), ceftazidime (CAZ; 30 µg) and aztreonam (ATM; 30 µg) respectively from the antibiotic resistance profiling were regarded as potential ESBL producers according to the Clinical and Laboratory Standards Institute (CLSI) criteria [37], and subjected to a confirmatory Double Disc Synergy Test (DDST). The DDST confirmatory test was carried out as described by Jarlier et al. [38] using two oxyimino-cephalosporins (CTR and CAZ) and a monobactam (ATM) all placed at 20 mm distance apart center to center from an AUG disc placed at the center. Positive DDST result is indicated by an enhancement of inhibition zone of the cephalosporins and monobactam towards the central augmentin disc [39, 40].

## **RESULTS AND DISCUSSION**

### **Presence of Foodborne Pathogens**

To assess the presence of selected bacterial foodborne pathogens, forty (40) hawked Kunun-zaki samples were examined for *Salmonella* spp and *S. aureus*.

*Salmonella* spp were detected in 19 (47.5%) out of the 40 Kunun-zaki samples examined (Figure 1), and the total *Salmonella* spp count (CFU/ml) ranged from  $1.90 \times 10^5$ – $9.70 \times 10^6$  (Table 1).

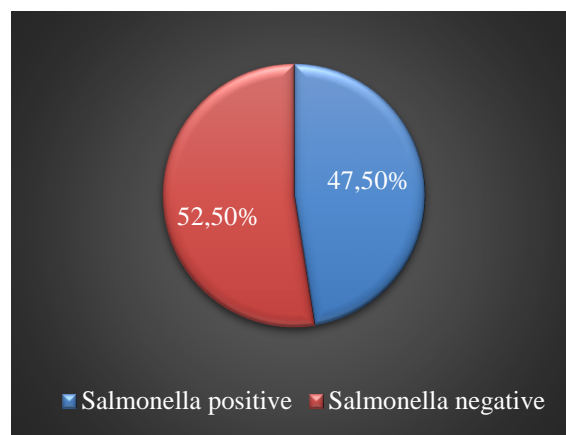


Figure 1 – Percentage occurrence of *Salmonella* spp isolates from the Kunun-zaki

Table 1 – The *Salmonella* spp count (CFU/ml) and *S. aureus* count (CFU/ml) in the Kunun-zaki samples

Kunun-zaki samples	<i>Salmonella</i> spp count (CFU/ml)	<i>S. aureus</i> count (CFU/ml)
KZ1	1.27×10 <sup>6</sup>	3.40×10 <sup>5</sup>
KZ2	-	4.50×10 <sup>5</sup>
KZ3	-	5.90×10 <sup>5</sup>
KZ4	8.30×10 <sup>6</sup>	3.70×10 <sup>5</sup>
KZ7	7.70×10 <sup>5</sup>	5.60×10 <sup>5</sup>
KZ8	8.00×10 <sup>5</sup>	-
KZ9	6.70×10 <sup>5</sup>	-
KZ10	-	6.40×10 <sup>5</sup>
KZ11	-	3.60×10 <sup>5</sup>
KZ12	-	5.90×10 <sup>6</sup>
KZ13	5.50×10 <sup>5</sup>	1.58×10 <sup>6</sup>
KZ14	-	6.40×10 <sup>6</sup>
KZ16	7.60×10 <sup>5</sup>	-
KZ18	1.90×10 <sup>5</sup>	-
KZ19	3.60×10 <sup>6</sup>	6.90×10 <sup>5</sup>
KZ20	7.40×10 <sup>5</sup>	-
KZ21	3.40×10 <sup>5</sup>	1.37×10 <sup>6</sup>
KZ22	-	7.50×10 <sup>5</sup>
KZ24	3.20×10 <sup>5</sup>	-
KZ25	-	6.80×10 <sup>5</sup>
KZ27	6.30×10 <sup>5</sup>	-
KZ28	-	7.40×10 <sup>5</sup>
KZ29	-	3.40×10 <sup>4</sup>
KZ30	9.10×10 <sup>5</sup>	6.50×10 <sup>5</sup>
KZ31	-	1.28×10 <sup>6</sup>
KZ33	1.12×10 <sup>6</sup>	-
KZ34	7.30×10 <sup>5</sup>	7.80×10 <sup>5</sup>
KZ35	7.40×10 <sup>5</sup>	1.46×10 <sup>6</sup>
KZ36	-	5.70×10 <sup>5</sup>
KZ38	1.78×10 <sup>6</sup>	-
KZ39	9.70×10 <sup>6</sup>	-
KZ40	-	7.50×10 <sup>6</sup>

Notes: KZ= Kunun-zaki sample

Previous reports have shown varying percentage rates of incidence. For instance, Gyar et al. [41] reported the recovery of *Salmonella* in 7 (28%) out of 25 Kunun-zaki samples examined in Keffi Metropolis, Nigeria; whilst Braide et al. [42] reported the recovery of *Salmonella* from 12 (75%) out of 16 Kunun-zaki samples bought and examined in Owerri, Nigeria. More so, Aboh and Oladosu [43] reported the recovery of *Salmonella* from 1 (2.4%) out of a total of 41 Kunun-zaki samples examined in Abuja, Northern Nigeria. The disparity in the number of *Salmonella* isolates may be due to the differences in the number of Kunun-zaki samples studied, the isolation procedures implemented and the cultural habit of the population; as well as discrepancies in the quality control

standards in the processing, preparation, storage, packaging and delivery of the Kunun-zaki.

According to the Center for Food Safety [44], there should be no detectable *Salmonella* spp in any mg/ml of any processed food substance ready and fit for human consumption. *Salmonella* spp are a significant cause of foodborne diseases and may colonize the intestinal tract of a large number of mammals and birds causing symptoms such as nausea, vomiting, abdominal cramps, diarrhoea fever, and headache [45].

Going forward, *S. aureus* was detected in 22 (55%) out of the 40 Kunun-Zaki samples (Figure 2), and the total *S. aureus* count (CFU/ml) ranged from 3.40×10<sup>4</sup>-7.50×10<sup>6</sup> (Table 1).

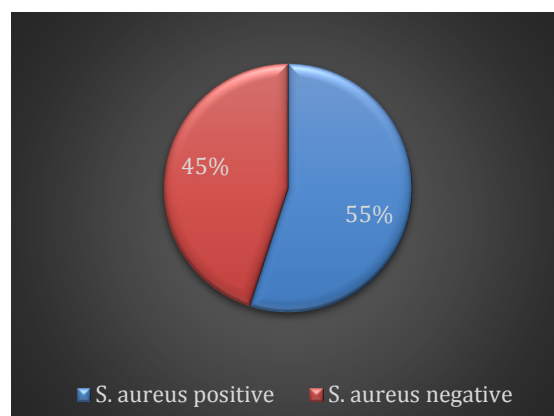


Figure 2 – Percentage occurrence of *S. aureus* isolates from the Kunun-zaki samples

Similar results have been reported in previous studies, although with a higher percentage rate of incidence. For instance, Gyar et al. [41] reported that 18 (72%) out of the total 25 Kunun-zaki samples examined in Keffi metropolis, Nigeria, were contaminated with *S. aureus*, whilst Braide et al. [42] reported the recovery of *S. aureus* from 11 (69%) out of the 16 Kunun-zaki samples examined in Owerri, Nigeria. Aboh and Oladosu [43] reported a much lower incidence rate in Abuja, Nigeria (6 (14.6%) *S. aureus* positive samples out of the total 41 Kunun-zaki samples examined).

Again, the discrepancies in the number of *S. aureus* isolates reported might be attributed to the differences in the number of Kunun-zaki samples examined, the isolation procedures implemented and probably, the cultural habit of the population; as well as the discrepancies in the quality control standards in the processing, preparation, storage, packaging and delivery of the Kunun-zaki.



According to the Center for Food Safety [44], the satisfactory limit for *S. aureus* detection in food is <20 CFU/ml with a borderline of 20 - ≤10<sup>4</sup> CFU/ml, above which the food is deemed unsatisfactory. Hence, the *S. aureus* count revealed that the Kunun-zaki drinks were unfit for consumption. *S. aureus*, as a foodborne pathogen has the potential to cause food poisoning or intoxication through the production of several enterotoxins.

The presence of *Salmonella* spp and *S. aureus* in Kunun-zaki samples might result from both direct contamination from the food handlers at the various stages of the unit operations; and indirectly from the water and containers used in its preparation and packaging, the substrates and ingredients used for preparation, and the environment where the Kunun-zaki samples are processed and hawked [46].

Furthermore, since the preparation process of Kunun-zaki involves pasteurization, a process that could eliminate both *Salmonella* spp and *S. aureus*, the other media through which the drinks can be contaminated would be the sugars, sweeteners and / or water added to the drink after cooling, just before packaging.

### Antibiotic Resistance

Modified Kirby Bauer disc diffusion test of the isolates revealed varying levels and patterns of susceptibility to the antibiotics tested (Figure 3 and Figure 4). The *Salmonella* spp were resistant to the beta lactam class of antibiotics used (IMP 58%, MEM 90%, AMP 53%) (Figure 3).

The *S. aureus* isolates revealed high resistance to oxacillin (41%) and the oxyimino-cephalosporin antibiotics used (CAZ 55%, CTR 41%) (Figure 4). Interestingly, 91% of the *Staphylococcus aureus* isolates were classified as multidrug resistant because they were MRSA and / or non-susceptible to at least one antimicrobial agent in three or more antimicrobial categories as defined by [47] (Table 2).

The persistent and indiscriminate exposure of bacterial strains to a multitude of antibiotics has led to the development of multidrug resistant bacteria [48]. Another possible medium for the development of MDR is through plasmid mediated resistance [49]. However, over 90% of the foodborne pathogens isolated from this study were susceptible to treatment with ofloxacin. This increased sensitivity to ofloxacin may stem from the fact that the antibiotics are not frequently used in therapeutic or prophylaxis therapy due to its high cost [49].

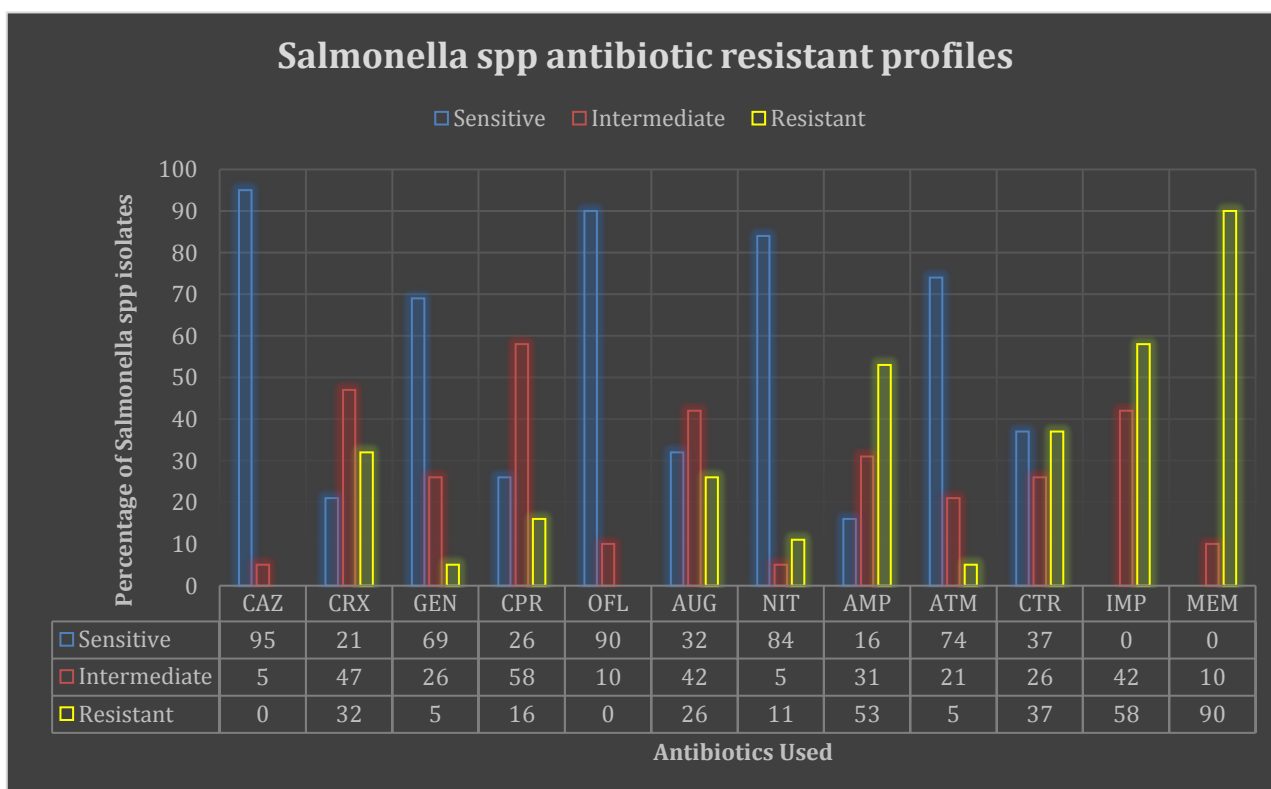


Figure 3 – Antibiotic-resistant profiles of the *Salmonella* spp isolates

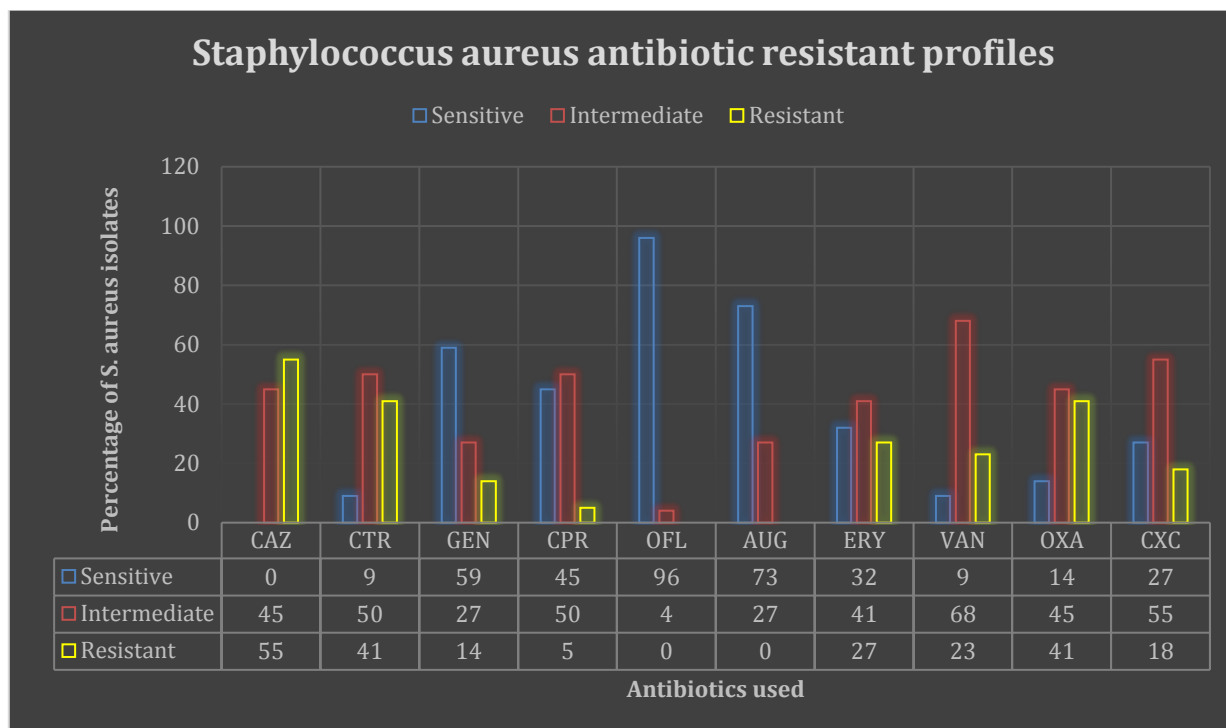


Figure 4 –Antibiotics resistant profiles of the *Staphylococcus aureus* isolates

Table 2 – Multidrug resistance in the *Staphylococcus aureus* isolates

<i>S. aureus</i> isolates	Antimicrobial category					MDR status
	Aminoglycosides (GEN)	Fluoroquinolones (CPR)	Macrolides (ERY)	Glycopeptides (VAN)	Anti-Staphylococcal Beta Lactams (OXA)	
KZ1	S	NS	S	NS	NS	MDR
KZ2	S	NS	S	NS	NS	MDR
KZ3	NS	S	S	NS	NS	MDR
KZ4	NS	S	NS	S	NS	MDR
KZ7	S	S	NS	NS	NS	MDR
KZ10	S	S	NS	NS	S	NO MDR
KZ11	S	NS	S	NS	NS	MDR
KZ12	S	S	NS	NS	NS	MDR
KZ13	S	S	S	NS	S	NO MDR
KZ14	S	NS	NS	NS	NS	MDR
KZ19	NS	NS	NS	NS	S	MDR
KZ21	S	S	NS	NS	NS	MDR
KZ22	NS	S	NS	NS	NS	MDR
KZ25	NS	NS	NS	NS	NS	MDR
KZ28	NS	NS	NS	NS	NS	MDR
KZ29	NS	NS	NS	NS	NS	MDR
KZ30	NS	S	S	NS	NS	MDR
KZ31	S	NS	NS	S	NS	MDR
KZ34	S	NS	NS	NS	NS	MDR
KZ35	S	NS	S	NS	NS	MDR
KZ36	NS	S	NS	NS	NS	MDR
KZ40	S	NS	NS	NS	NS	MDR
MDR Incidence Percentage						91%

Notes: S=susceptible, NS=non-susceptible

With regards to antibiotic resistance, MRSA, VRSA and ESBL producing *Enterobacteriaceae* are currently of particular public health concern. In this study, 41% of the isolates were MRSA, 23% were

VRSA and 68% were VISA (Figure 4). In addition, 26% of the *Salmonella* spp isolates were confirmed to be ESBL producers based on the DDST (Figure 5).

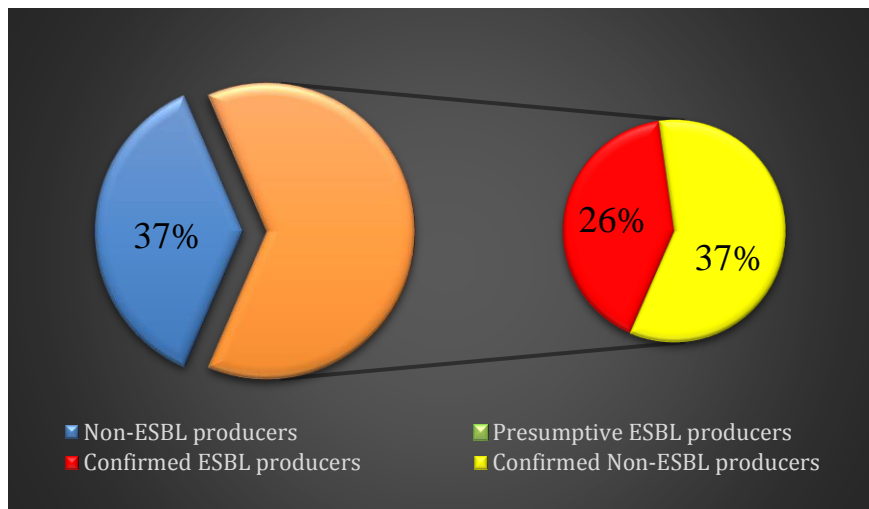


Figure 5 – Percentage occurrence of ESBL producing *Salmonella* spp based on the phenotypic breakpoint and Double Disc Synergy Test (DDST)

Previously, vancomycin has been widely utilized for the treatment of MRSA infections and this has led to the emergence of VRSA and VISA [25]. Following treatment of MRSA patients with vancomycin, VISA isolates emerged as a result of mutations in MRSA isolates [50].

## CONCLUSION

The results obtained from this study revealed that Kunun-zaki sold in Umuahia metropolis, Nigeria, is contaminated with important bacterial food-borne pathogens, and whose counts do not conform to the standard limits of detectable microbes in ready-to-eat foods as stipulated by the Center for Food Safety. Hence, the consumption of these

products may constitute serious public health concerns if not correctly quality controlled. From this study, it is evident that this product could include a source of ESBL producing *Salmonella*, MRSA and VRSA, amongst other pathogenic microbes which were not detected in the study but have been previously reported. Whilst consumption of Kunun-zaki is encouraged due to its health and nutritional benefits as registered from recent and previous studies, its production, processing and packaging should be regulated appropriately, and strict compliance with acceptable hygiene practices during any step of preparation, storage and distribution to the final consumer are essential to prevent contamination with foodborne pathogens and any health complications that may result thereof.

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