

# Detection and Antibigram of Bacterial Contaminants in Some Commonly Consumed Indigenous Street Vended Foods in Kasua Central Market in Kaduna North, Kaduna State, Nigeria

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## Abstract

Some indigenous ready-to-eat foods, due to its affordability and availability serve as a major alternative to restaurants and high profiled eateries in developing countries. The hygiene standard of these foods is a critical point that needs to be considered due to microbial contamination by potential pathogens. A triplicate of three different food samples: Fura, Nunu, and Tuwo were collected and analysed to determine their microbiological safety. Bacterial isolates were identified using standard methods while antibiotic sensitivity test and multiple drug resistance pattern were carried out. The isolated bacteria: *Escherichia coli*, *Klebsiella pneumoniae*, *Salmonella 100yphi*, *Staphylococcus aureus*, and *Enterobacter sp.* While fungal isolates include: *Aspergillus niger*, *Aspergillus flavus*, *Rhizopus. oryzae* and *Penicillium sp.* *Salmonella sp.* was observed to be resistant to 70% of the antibiotics while both *Klebsiella sp.* And *Enterobacter sp.* exhibited resistance to 60% of the tested antibiotics. *S. aureus* had the least resistance to about 30% of the tested Gram-positive antibiotics. Out of all the tested antibiotics, only Tarivid was observed to be effective against all the Gram-negative bacterial isolates, while Reflacine and Ciprofloxacin were effective against 80% of them. The presence of coliform bacteria in the sampled foods raise a great concern about the safety of the street vended foods in the sampled region in Nigeria. The microbiologically low quality of the vended ready-to-eat foods assayed in this study might have resulted from a relatively low level of hygiene observed by the vendors, as well as absence of regulatory body or hygiene codes to guide the street food vendors. From the results of this study, efforts should be intensified by all concerned to make sure that the safety of people remains paramount in the ways the vended foods are produced and distributed.

**Keywords:** Street vendors, Bacteria, Multidrug resistance pattern; Antibiotics, Hygiene.

## INTRODUCTION

Street vending and hawking of foods provide income and job as well as social roles in developing countries. Vending of foods on the streets records high patronage due its affordability, and availability. Ready-to-eat foods require no further processing or cooking, and it is majorly prepared and served in streets and local public places (Rakha *et al.*, 2022).

The practice of food vending in Nigeria plays an important role in meeting the food needs of urban residents, but with associated numbers of health risks (Chukuezi, 2010). Concerns have been raised regarding the safety and hygiene standards in roadside food processing (Okoché *et al.*, 2015). Poor construction and sales locations as well as lack of facilities such as garbage collection points have been described as the main cause of poor food safety and quality. Street vended foods are often processed and sold at unsafe conditions such as sales locations which

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include kiosks, make-shift housing, push carts and other temporary structures, as well as inaccessibility to running water at vending sites (Osalumhense and Ekundayo, 2021). Since there are no strict regulations of ensuring the safety of street vended foods. The vendors practice poor personal hygiene and as such serve as potential carriers of pathogens which contaminate food materials during handling or processing (Nnachi *et al.*, 2014).

The World Health Organization (WHO) (2012) reported that low- and middle-income countries, especially those in the African and Southeast Asian sub-regions, suffer significantly from the burden of foodborne diseases (Havelaar *et al.*, 2015), sometimes resulting in high economic costs. According to the latest World Health Organization (WHO) report, more than 91 million people are affected in Africa. WHO (2015) reported that over 2.2 million deaths of children were caused by food-borne diarrhoea annually due to the consumption of unhealthy food. Majority of the cases of food poisoning each year have been found to be associated with contamination by *Staphylococcus aureus*, *Salmonella*, *Clostridium perfringes*, *Clostridium botulinum*, *Campylobacter sp*, *vibro parahaemolyticus*, *Bacillus cereus*, enteropathogenic *Escherichia coli* as well as some fungi and the contamination of food materials is due to secretion of toxic compounds such as cereulide by *B. cereus* and mycotoxins (Fadahunsi and Makinde, 2018; Gdoura-Ben *et al.*, 2018).

The poor sanitary conditions of many local markets and their highly polluted, chaotic and pathogenic flora-infested environments are the most common causes of contamination of food sold by such street vendors (Adesemoye *et al.*, 2021). Poor hygiene in food processing environments is known to contribute to the spread and contamination of foodborne pathogens (Zhao *et al.*, 2017). Contaminated working surfaces during food processing are a major source of contamination in street vended foods. Poor personal hygiene can result in food contamination for example when food personnel fails to wash hands properly after using the toilet, is a serious risk of faecal contamination (Ismail *et al.*, 2016). The lack of proper monitoring of street food vendors or hygiene codes for the vendors limits the consciousness of the vendors to constant hand washing during food processing. Street vended foods are mostly patronized by low-income earners due to its low cost and affordability and this low cost is sometimes achieved through the use of sub-standard raw materials- like tomato, pepper, meat- by the vendors, that can serve as source of microbial contamination (Sezgin and Şanlıer, 2016).

Since microbial food contamination may emanate from contaminated processing equipment or cooking utensils to food, microbiological examination of foods and contact surfaces during processing, as well as the sanitary conditions under which the food is processed, will provide information concerning the quality of the foods from local vendors (Adolf and Azis, 2012). It is good to note that many foodborne illnesses can be prevented through proper handling and storage of food. Stakeholders in street food trade, especially street food vendors, consumers and governments, need to be involved in ensuring the safety of the food sold (Alimi, 2016). This study evaluated the bacterial contaminants of common street vended foods in Kasua central market in Kaduna North Kaduna State, Nigeria by isolating pathogenic microorganisms that may pose threat to the health of consumers.

## MATERIALS AND METHODS

### Sampling of Foods

Fura, Nunu, and Tuwo were collected from street food vendors in Kasua central market in Kaduna North, Kaduna State, Nigeria. Three (3) samples were collected from different locations in Tudun Wada at 10. 519647° N , 7. 428183° E . Samples were collected into sterile sample bottles and packaged in zip-lock bags, and then transported to the microbiology laboratory for analysis.

### Isolation and Identification of Bacterial Contaminants in the Sampled Foods

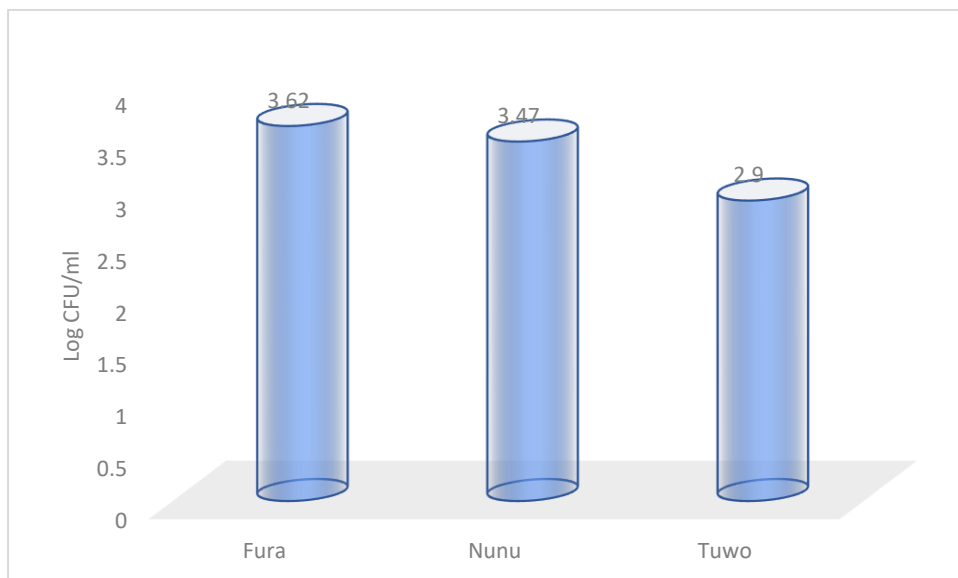
The culture media (MacConkey agar and Nutrient Agar) were prepared according to manufacturer's standards and then sterilised in an autoclave at 121° C and 15 psi for 15 minutes. A seven-fold serial dilutions of the food samples were prepared aseptically using normal saline. One (1) mL of the dilutions 7, 6, and 4 were aseptically inoculated into labelled sterile Petri plates. The sterilised agar media were allowed to cool. The media were then dispensed into the Petri plates. The experiment was repeated in triplicate. The media were allowed to set and the bacterial plate cultures were incubated at 37 °C for 24 hours. The bacterial loads were estimated and distinct colonies were sub-cultured to obtain pure isolates. The pure cultures were characterised morphologically using Gram and spore staining and biochemically with catalase, coagulase, oxidase, indole, citrate utilisation, and sugar fermentation tests carried according to Adesetan *et al.* (2017), to ascertain the probable identity of the bacterial isolates.

### Antibiotics Sensitivity Test

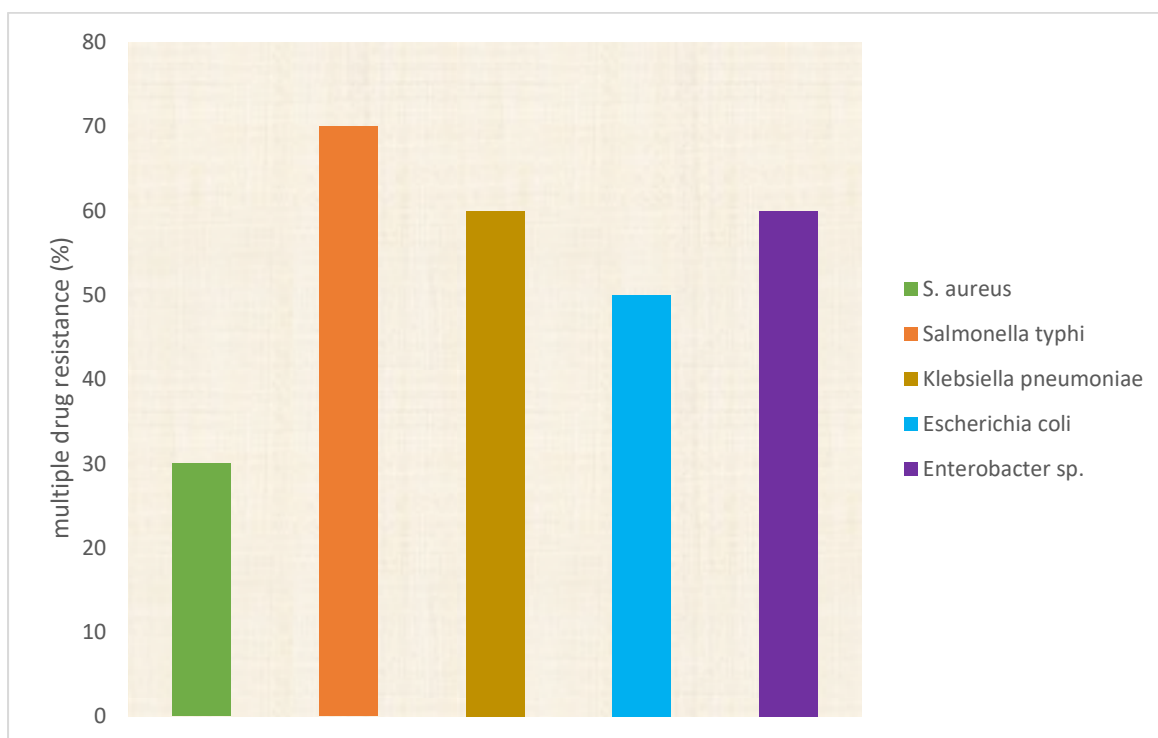
A 24-hour broth culture of the bacteria isolates was prepared and standardized using 0.5 McFarland standard. With the aid of sterile swab stick, each standardized inoculum was aseptically inoculated on freshly prepared Mueller Hinton agar using streaking method. Antibiotics discs were aseptically placed on each Petri plate based on the Gram reaction of each isolate. The plates were then incubated at 37 °C for 24 hours. The multidrug resistant pattern of each isolate was observed as well as the inhibitory efficacy of each antibiotic against all the bacterial isolates (Onanuga and Temedie, 2011).

## RESULTS

The results in figure 1 revealed that the samples were contaminated more than the others. The values of 3.62, 3.47 and 2.9 Log CFU/mL were isolated from Fura, Nunu and Tuwo respectively. The morphological attributes of the bacterial isolates varied in colour, shape, margin, elevation and surface appearance (Table 1). Biochemical characterization of the isolates indicated that they belong to *Escherichia coli* (2), *Klebsiella pneumoniae* (2), *Salmonella typhi* (2), *Staphylococcus aureus* (2) and *Enterobacter* sp. (1) (Table 2). In table 3, the Gram-positive bacterial isolate was observed to be sensitive to six of the antibiotics used while resistant to one. Among the Gram-negative bacteria, all the isolates were sensitive to OFX, resistant to AU, CEP, NA and AMP while their resistance to other antibiotics used in this study varied (Table 4). Figure 2 depicts multiple drug resistance to the antibiotics. The resistance pattern showed that *Staphylococcus aureus* < *E coli* < *Klebsiella pneumoniae* < *Enterobacter* sp. < *Salmonella typhi* while their sensitivities to tested antibiotics are in the range of OFX > PEF > CPX > CN > S > SXT (Figure 3).



**Figure 1:** Population density of bacterial isolates form food samples



**Figure 2:** Multiple drug resistance pattern of the bacterial isolates

**Table 1: Morphological Attributes of Bacterial Isolates**

Sample	Colour	Shape	Margin	Elevation	Surface
FRA1	White	Circular	Lobate	Raised	Smooth
FRA2	Cream	Circular	Lobate	Raised	Smooth
FRA3	Colourless	Circular	Undulating	Low convex	Smooth
FRA4	Yellow	Circular	Lobate	Convex	Smooth
NNU1	Cream	Circular	Lobate	Flat	Smooth
NNU2	Cream	Circular	Lobate	Raised	Smooth
NNU3	Colourless	Circular	Undulating	Low convex	Smooth
NN4	White	Circular	Lobate	Raised	Smooth
TUW1	Yellow	Circular	Lobate	Convex	Smooth

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**Table 2: Biochemical characteristics of bacteria obtained from different food samples**

Sample	Shape	Grams reaction	Catalase	Coagulase	Motility	Citrate	Indole	Spore staining	Oxidase	Fermentation test			Bacteria isolated
										Glucose	Fructose	Lactose	
FRA1	Rod	-	+	-	+	-	+	-	-	+	-	+	<i>Escherichia coli</i>
FRA2	Rod	-	+	-	-/+	+	-	-	-	+	+	-/+	<i>Klebsiella pneumoniae</i>
FRA3	Rod	-	+	-	-	-	-	-	-	+	-	-	<i>Salmonella typhi</i>
FRA4	Cocci	+	+	+	-	+	-	-	-	+	+	+	<i>Staphylococcus aureus</i>
NNU1	Rod	-	+	-	+	+	-/+	-	-	+	+	+	<i>Enterobacter sp</i>
NNU2	Rod	-	+	-	+	+	-	-	-	+	+	+	<i>Klebsiella pneumoniae</i>
NNU3	Rod	-	+	-	+	-	-	-	-	+	-	-	<i>Salmonella typhi</i>
NNU4	Rod	-	+	-	+	-	+	-	-	+	-	+	<i>Escherichia coli</i>
TUW1	Cocci	+	+	+	-	+	-	-	-	+	+	+	<i>Staphylococcus aureus</i>

Key: Positive = +; Negative = -; Variable = -/+

**Table 4: Antibiotic sensitivity test of Gram-positive bacterial isolates**

Bacteria isolated	Antibiotics sensitivity										
	CPX	NB	CN	AML	S	RD	E	CH	APX	LEV	
<i>Staphylococcus aureus</i>	S	R	S	0	0	S	S	0	S	S	

Key: CPX: Ciproflox, NB: Norfloxacin, CN: Gentamycin, AML: Amoxil, S: Streptomycin, RD: Rifampicin, E: Erythromycin, CH: Chloramphenicol, APX: Ampiclox, LEV: Levofloxacin.  
S= sensitive; R= resistance

**Table 5: Antibiotic sensitivity of Gram-negative bacterial isolates**

	Antibiotics sensitivity										
	OFX	PEF	CPX	AU	CN	S	CEP	NA	SXT	AMP	
<i>Salmonella typhi</i>	S	S	S	R	R	R	R	R	R	R	
<i>K. pneumoniae</i>	S	R	R	R	I	I	R	R	I	R	
<i>Escherichia coli</i>	S	S	S	R	S	S	R	R	R	R	
<i>Escherichia coli</i>	S	S	S	R	R	I	R	R	S	R	
<i>Enterobacter sp.</i>	S	S	S	R	S	R	R	R	R	R	

Key: OFX: Ofloxacin, PEF: Pefloxacin, CPX: Ciprofloxacin, AU: Augentin, CN: Gentamycin, S: Streptomycin, CEP: Ceporex, NA: Nalidixic acid, SXT: Septrin, AMP: Amplicin.  
S= sensitive; R= resistance ; I= intermediate

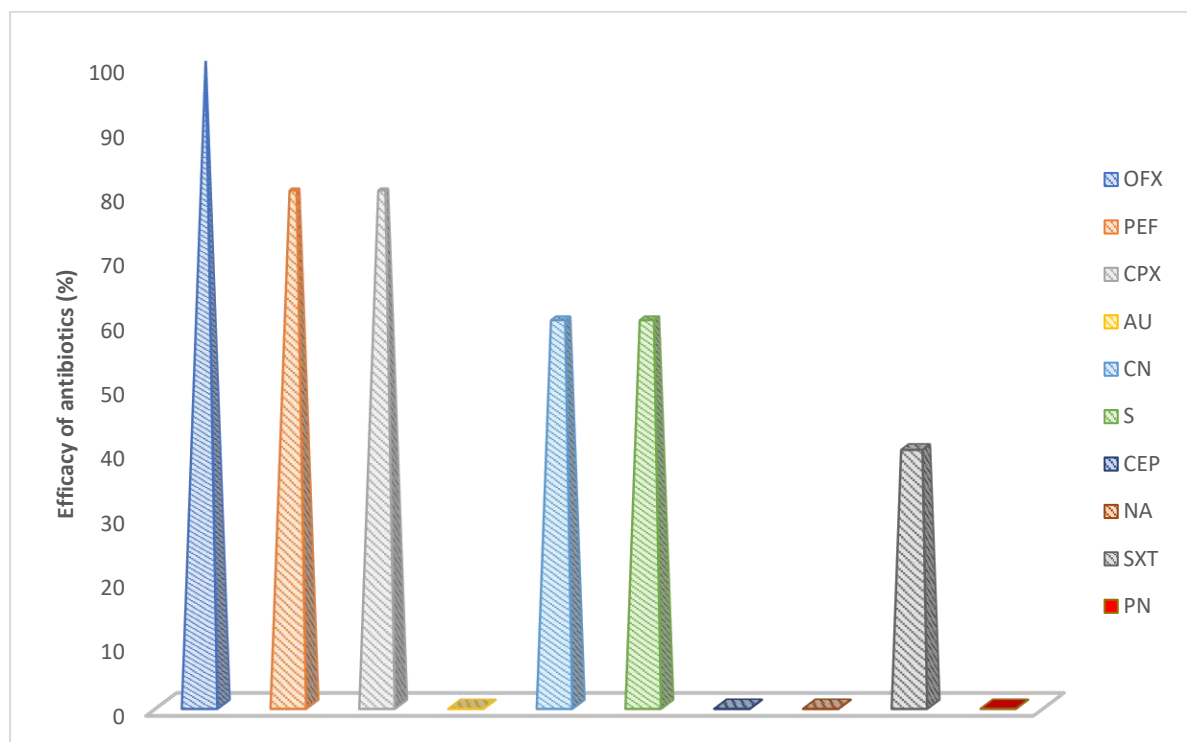


Figure 3: The sensitivity of tested antibiotics against the bacterial isolates

## DISCUSSION

Majority of the bacteria contaminants that impact food quality and safety are either introduced during the processing and handling or from the raw materials (Adesemoye *et al.*, 2021). For bacteria to truly cause an infection, certain number of the pathogen known as infective dose (ID<sub>50</sub>) must be present or ingested by the host. From this study, *Fura* which is usually produced from millet or sorghum was observed to have the highest microbial load while *Tuwo* which is usually made from rice or corn was observed to have the least bacterial load. The load observed in this study was 10<sup>4</sup> CFU/mL is similar (although slightly higher) to the findings of Agboola *et al.* (2018) who detected bacterial load of 1.3 × 10<sup>3</sup> CFU/mL in the sampled vended food materials in Ibadan. The report of Amare *et al.* (2019), however is in agreement with the findings of this current study; as 10<sup>4</sup> CFU/mL was recorded as bacterial load. Alem (2020), reported a total bacterial load of 10<sup>3</sup> CFU/mL in tested street vended foods. A 10<sup>6</sup> CFU/mL bacterial load was also reported by Adesetan *et al.* (2017) from street vended foods.

Majority of the isolates in this study are known human pathogen; *E. coli*, *Klebsiella*, *Salmonella*, *Enterobacter*, and *Staphylococcus* sp. The presence *E. coli*, *Staphylococcus aureus* and *Salmonella* sp. and *Klebsiella* sp. in street vended foods in various parts of Nigeria has earlier been reported by different researchers (Fadahunsi and Makinde, 2018; Alem, 2020; Adesemoye 2021). *K. pneumoniae* is often reported in foods such as vegetables, fish, meat, etc. in recent times, there has been an increase in the number of food-borne outbreaks caused by this pathogen (Davis and Price, 2016). Many virulent factors including capsules, endotoxins, adhesins, iron-scavenging systems, and siderophores have been identified in *Klebsiella* sp. (Kang *et al.*, 2015). Although *Klebsiella pneumoniae* is considered an opportunistic pathogen, it produces large amounts of endotoxin that can cause fatal infections (Katty *et al.*, 2022). The presence of *Staphylococcus* sp. in food indicates poor personal hygiene and inappropriate handling and manufacturing practices by food processor (Aung and Chang, 2014). *Staphylococcus* sp produces enterotoxins that can withstand high temperatures and can cause

vomiting and diarrhoea if ingested. It is also resistant to high concentrations of sodium chloride (Lorenzo *et al.*, 2018). Deaths from staphylococcal food poisoning are rare (Fusco *et al.*, 2015), but can be fatal in young children and immunocompromised individuals.

The presence of *E. coli* in vended foods is becoming recurrence issue. This pathogen has been isolated from food samples by several researchers. Were *et al.* (2020) and Hariri, (2022) reported isolated this pathogen from street vended foods, chicken, beef and fish. *Salmonella typhi*, a notable pathogenic bacteria has also been isolated from vended foods by other researchers (Hungaro *et al.*, 2013; Anukampa *et al.*, 2017). Amare *et al.* (2019) reported isolation of this pathogen from street vended foods. The bacterium is becoming more frequent in most street vended foods, raising a lot of public health concerns (Putturu *et al.*, 2015). *Salmonella typhi* is a well-known pathogen of both human and animal; and its pathogenesis include production of inflammatory cytokines by stimulating the mucosa of epithelial cells (Ahmad *et al.*, 2022). The genus *Enterobacter* is also one of the commonly found pathogens in foods and quite a lot of studies have linked this pathogen to foodborne gastroenteritis which usually results from poor environmental sanitation and personal hygiene (Igbinosa *et al.*, 2020). Although some species of *Enterobacter* are commonly found in the gut as normal flora, several species have been implicated in infections such as wound infection, urinary tract infection, nosocomial infection, endocarditis, diarrhoea, etc. (Dougnon *et al.*, 2020)

### **Antibiotic Sensitivity**

The bacteria isolated in this study were subjected to antibiotic sensitivity test and it was observed that *Staphylococcus aureus*. was very sensitive to nearly all the tested antibiotics with exception of Amoxil, Streptomycin and Chloramphenicol; As a result, the isolated *Staphylococcus aureus* had the lowest number of multiple drug resistance pattern while other isolated bacteria had higher number of drug resistance pattern between 5 and 7; with *Salmonella* sp. having the highest number of drug resistance, while both *Enterobacter* sp. and *Klebsiella* sp. were resistant to six (6) of the tested antibiotics. In this study, multidrug resistance was taken as resistance to three or more antibiotics as all the isolated bacteria were observed to have multiple drug resistance. All the isolated Gram-negative bacteria from this study were resistant to Augmentin, Ceporex, Nalidixic acid, and Ampicilin. However, they were slightly sensitive to Gentamycin and Streptomycin. Tarivid, Reflacine and Ciprofloxacin were very active against the isolated Gram-negative bacteria, with the exception of *Klebsiella* sp. which was resistant to both Reflacine and Ciprofloxacin, but was sensitive to Tarivid. Out of the tested antibiotics in this study, only Tarivid was found to be 100% effective against all the tested bacteria isolates, while PEF and CPX showed 80% efficacy. Multiple drug resistance in *Salmonella* has been known to be highly prevalent in recent times. According to Mina *et al.* (2023), about 85% of the tested *Salmonella* sp. showed resistance to the tested antibiotics. The opportunistic *Staphylococcus aureus* known to be a normal flora of the skin, can cause a variety of pyogenic and systemic infections due its possession of virulence factors such as adhesins, enzymes, toxins, and immune evasion proteins (Sonola *et al.*, 2021). Onanuga and Temedie (2011), reported that intestinal *Staphylococcal* infection was highly resistant to antibiotics and multiple drug resistance pattern was also observed in the tested *Staphylococcus* isolates. Wartu *et al.* (2019) observed that many of the drug resistance observed in bacteria in recent time can be traced to extrinsic factors. Many of the pathogens developed ways to survive in the presence of the antibiotics due to unscrupulous use and abuse of the currently available antibiotics (Ferreira *et al.*, 2019). Ferreira *et al.* (2019), attributed the multidrug resistance reported in *Klebsiella* sp. to the presence of several antibiotic resistance genes most especially, extended spectrum beta-lactamases (ESBL)-encoding genes. The

presence of efflux pump, and porin-coding genes also contribute to drug resistance in the bacteria.

The presence of coliform bacteria in the sampled foods in this study raises a great concern about the safety of the street vended foods in the Northern parts of Nigeria. The reports of Agboola *et al.* (2018) from sampled foods in different parts of Nigeria, as well as reports from other researchers suggests that street vended foods in many parts of Nigeria are of low quality microbiologically; and as such raise a public health concern. Over-prescription and indiscriminate abuse of some of these antibiotics has been listed as one of the major contributors to multiple drug resistance pattern in most pathogenic bacteria (Akinyele *et al.*, 2017).

## CONCLUSION

Street vended foods are a great succour to both the vendors and the customers. The microbiological quality and safety of the many foods vended on the streets remains a serious concern, because data from many researches, including this current study have isolated pathogenic bacteria (including coliforms) from these foods. Hence there should be concerted efforts by all concerned to make sure that the safety of people remains paramount in the ways the vended foods are produced and distributed. Strengthening food safety policies and strict enforcement of food safety regulations can drastically reduce the risk of food-borne illness to safe levels.

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