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Research Article

Prevalence of Bacterial Loads on some Fruits and Vegetables Sold in Kaduna Central Market, Northwestern Nigeria

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Abstract

Background and Objective: Fruits and vegetables served as the major sources of plant proteins, vitamins and fibers that support human health. This study was conducted to assess the bacteriological quality of various fruits and vegetables sold in Kaduna central market, northern Nigeria. **Materials and Methods:** Samples of the fruits and vegetables were procured from the market and analyzed using serial dilution technique and inoculation was done on MacConkey and nutrient agar and incubated for 48 h. **Results:** The result obtained showed the presence of six bacterial species: *Staphylococcus aureus*, *Streptococcus* sp., *Enterobacter* sp., *Escherichia coli*, *Citrobacter* sp. and *Klebsiella* sp. *Staphylococcus aureus* was the most abundant (with 80% relative occurrence) while *Streptococcus* sp. was the least abundant (with 2% relative occurrence). **Conclusion:** It was concluded that 6 different bacterial species prevailed on the fruits and vegetables sold in Kaduna central market: The result therefore implied that people consuming these fruits and vegetables are at a higher risk of pneumonia and toxic shock syndrome due to Staphylococcal infection, strep throat, rheumatic fever, scalded skin syndrome, scarlet fever and puerperal fever due to *S. aureus*, gastrointestinal disorders due to *E. coli* and bronchopneumonia due to *Klebsiella* sp. There is therefore; the urgent need for orienting the general populace on the inherent dangers associated with consumption of these types of fruits and vegetables without thorough disinfection.

Key words: Bacteria, fruits, infection, *Klebsiella* sp., vegetables, toxic shock syndrome, Staphylococcal infection

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Fruits and vegetables are vital sources of nutrient to human beings. They give the body the necessary vitamins, fats, minerals and oil in the right proportion for human growth and development¹. However, the existence of fruits and vegetables is threatened by various factors such as changes in climatic condition, pest, inadequate rainfall and the activities of different bacterial communities. These bacterial biofilms cause spoilage and associated clinical symptoms. Spoilage is defined by Tournas² as any change in the condition of food in which the food becomes less palatable or even toxic; these changes may be accompanied by alterations in taste, smell, appearance or texture. The colonization process involves the ability of the bacteria to establish itself within the host. This is initiated when bacteria (following adhesion and release of enzymes) depolymerises certain specific cell wall polymers (such as proto pectin, the cementing substance) of the produce³. Susceptibility of fruits and vegetables is largely due to differential chemical composition such as pH and moisture contents. The higher pH (near neutrality) and moisture contents are associated with their greater predisposition to bacterial activities. The occurrence of bacterial species on fruits and vegetables is recognized as a source of potential health hazard to man and his animals. This is due to their production of bacteriotoxins compounds which are capable of inducing several critical clinical symptoms in man following ingestion or inhalation; even though they differ in their degree and manner of toxicity. These microbes found in produce may have other, less direct, impacts on human health. Exposure to non-pathogenic microbes associated with plants may influence the development of allergies and the consumption of raw produce may represent an important means by which new lineages of commensal bacteria are introduced into the human gastrointestinal system⁴. The contamination of fruits and vegetables by bacteria could also be as a result of poor handling practices in food supply chain, storage conditions, distribution, marketing practices and transportation⁵. This study therefore, aimed at investigating the prevalence and occurrence of different bacterial communities on the fruits and vegetables sold in Kaduna central market with the view of identifying the different species of bacteria with their potential toxigenicity. The results of this research was also intended to be used in suggesting possible ways of minimizing or avoiding possible health problem associated with these bacterial species.

MATERIALS AND METHODS

The samples were collected from Kaduna central market (Kaduna north local Government area), located along Ahmadu Bello Way, Kaduna state, Nigeria (lat 10°31'06.29"N and long 7°25'36.57"E). They include: Pepper (*Capsicum annum* L.), Tomato (*Lycopersicum esculentum* Mill.), Irish potato (*Solanum tuberosum* L.), Garden egg (*Solanum melongena* L.), Carrot (*Daucus carota* L.), Cucumber (*Cucumis sativus* L.), Water melon (*Citrullus lanatus* Thunb.), Onion (*Allium cepa* L.), Sweet potato (*Ipomoea batatas* L.) and Okra (*Abelmoschus esculentus* L.). The samples were collected twice monthly between April, 2017 to August, 2017 from whole seller and retailer, put into a clean polythene bag and labelled, respectively. The samples were kept in the refrigerator at 4°C for later use.

Bacterial isolation from fruits and vegetables: The methodology of Dashwood *et al.*⁶ and Balali *et al.*⁷ were used in the isolation of microbial flora from the fruits and vegetables. The surface were sterilized with 0.1% Mercury chloride (HgCl) for 2 min then rinsed three times with distilled water. A sterile blade and forceps were use to cut small section of the tissue containing both the healthy and the rotten portion. The cut portions were pounded with pestil and mortar to make paste.

Inoculation and incubation: Streaking method was used for inoculation of the organisms on different agar plates (MacConkey Agar, Mannitol Salt Agar, Salmonella-Shigella Agar and Blood Agar). The pulverized sample was transferred to the edge of an agar plates with a sterilized wire loop and then a parallel, non-overlapping streaks were made on the surface of the already solidified agar plates. Streaked plates were incubated in the incubator for⁸ 24 h at 37°C.

Characterization of bacterial isolates: The various colonies observed in the plates were distinguished on the basis of their cultural characteristics such as colony size, shape, color, consistency and haemolytic characteristics as described by Fawole and Oso⁹. Bacterial growth was sub-cultured on NA and was preserved in NA slants. The representative bacterial colonies distinguished on the basis of their cultural characteristics were smeared thinly on grease free microscopic slide and Gram-stained. The stained smears were then examined microscopically under the oil immersion objectives.

The organisms recovered from the deteriorated vegetables were grouped on the basis of their Gram-reaction, cell morphology and cell arrangement. Isolates were further characterized using biochemical test described by Faiers *et al.*¹⁰ and Wreghitt and Morgan-Capner¹¹.

Statistical analysis: The data obtained were subjected to one way analysis of variance and a significance test for differences between sample variances using the least significance difference (LSD) in the comparison of means at 5% level of significance.

RESULTS

Biochemical characterization: The results of the biochemical characterization of Gram-positive bacterial biofilms on the vegetables in central Market Kaduna was presented in Table 1. The result showed that *Staphylococcus aureus* was the most prevalent Gram-positive bacteria while *Streptococcus* spp. was the least abundant. Similarly, biochemical characterization of Gram-negative bacterial isolates from the vegetables indicated that *E. coli*, *Klebsiella* spp. and *Citrobacter* are the major Gram-negative bacterial populations found associated with the fruits and vegetables in central market, Kaduna (Table 2).

Prevalence of the isolates: The result for the relative abundance of different bacterial species in the fruits and vegetables was presented in Table 3. The result showed that *S. aureus* had the highest occurrence with 80% while *Streptococcus* spp., has the least with 2%.

DISCUSSION

Fresh fruits and vegetables are extraordinary dietary sources of nutrients, micronutrients, vitamins and fibers for human and an essential basic material for the food industry. They are widely exposed to microbial contamination through contact with soil, dust and water. Handling at harvest and post harvest processing has also been established to cause spoilage and loss of quality. Fruits and vegetables harbor a wide range of microbial contaminants as reported by Long *et al.*¹². The major bacterial populations that are generally present on fruits and vegetables include species of *Pseudomonas* spp., *Bacillus* spp., *Enterobacter* spp., *Sarcina* spp., *Staphylococcus* spp., *Streptococcus* spp., *Lactobacillus* spp. and *Leuconostoc* spp. as stressed by Erin¹³. Most of the vegetables were not really fresh as they stayed in the market

Table 1: Biochemical characterization of Gram-positive bacterial in vegetables and fruits from Kaduna

Vegetables	Man							DNA	Inference
	Hem	Coa	O	F	Bil	Cat			
Tomatoes	A	+	+	-	-	+	+	<i>S. aureus</i>	
Cucumber	A	+	+	-	-	+	+	<i>S. aureus</i>	
Potato	A	+	+	-	-	+	+	<i>S. aureus</i>	
Onion	-	-	-	-	-	-	+	<i>Stapp</i> spp.	
Carrot	A	+	+	-	-	+	+	<i>S. aureus</i>	
G/egg	A	+	+	-	-	+	+	<i>S. aureus</i>	
S/potato	A	+	+	-	-	+	+	<i>S. aureus</i>	
Okro	B	-	-	-	+	+	-	<i>Strep</i>	
Pepper	A	+	+	-	-	+	+	<i>S. aureus</i>	
W/melon	A	+	+	-	-	+	+	<i>S. aureus</i>	

Hem: Haemolysis reaction, Coa: Coagulase reaction, DNA: Deoxyribonucleic acid, Man: Mannitol fermentation, Bil: Bile solubility, Cat: Catalase, +/-: Partial positive

Table 2: Biochemical characterization of Gram-negative bacteria on vegetables and fruits from Kaduna

Vegetables	Ind	MR	VP	CIT	MOT	Urea	S/F	Inference
Tomatoes	+	+	-	-	+	-	K/AG	<i>E. coli</i>
Cucumber	-	-	-	+	+	-	K/A	<i>Klebsiella</i> spp.
Potato	+	+	-	-	+	-	K/AG	<i>E. coli</i>
Onion	-	-	+	+	+	-	K/A	<i>Klebsiella</i> spp.
Carrot	-	-	+	+	+	-	K/A	<i>Enterobacter</i> spp.
Garden egg	-	-	-	+	+	-	K/A	<i>Klebsiella</i> spp.
Sweet potato	+	+	-	-	+	-	K/AG	<i>E. coli</i>
Okro	-	+	-	+	+	+	K/AGH ₂ S	<i>Citrobacter</i> spp.
Pepper	-	+	-	+	+	+	K/AGH ₂ S	<i>Citrobacter</i> spp.
Water melon	-	+	-	+	+	+	K/AGH ₂ S	<i>Citrobacter</i> spp.

Ind: Indole, MR: Methyl red, VP: Voges proskauer, Cit: Citrate test, Mot: motility, Urea: urea, S/F: Sugar fermentation

Table 3: Abundance of different bacterial species in the fruits and vegetables from kaduna central market

Bacterial isolates	Frequency	Percentage
<i>Escherichia coli</i>	2	04.00
<i>Staphylococcus aureus</i>	40	80.00
<i>Streptococcus</i> spp.	1	02.00
<i>Enterobacter</i> spp.	3	06.00
<i>Klebsiella</i> spp.	2	04.00
<i>Citrobacter</i> spp.	2	04.00
Total	50	100

for long periods resulting to their spoilage¹⁴. More so, most of the vegetables were grown under irrigation water containing enteric bacteria, viruses, protozoa or helminths, which subsequently increases the probability of the isolation of pathogens from harvested produce as reported by Uzeh *et al.*¹⁵.

The extent of spoilage depends on micro-organisms involved. Uzeh *et al.*¹⁵ reported that *S. aureus* was found in carrots, cucumber, cabbage and lettuce at food outlets within Lagos Metropolis. The presence of *Staphylococcus* and *Streptococcus* species in the vegetables observed in this study agrees with the work of Erin¹³ who reported the presence of

Staphylococcus spp. in pawpaw, orange and kola nut at Sango Market, Ilorin. However, Wells and Butterfield¹⁶ reported *Salmonella*, *Shigella* and gastro-intestinal viruses as the major pathogens associated with food poisoning in England and the United States. The presence of other bacterial species of *E. coli*, *Enterobacter* spp., *Citrobacter* spp. and *Klebsiella* spp. in the vegetables obtained from Kaduna central market conforms to the findings of Rangel *et al.*¹⁷ in which they reported the presence of *E. coli* in lettuce, apple, salads, coleslaw, melons, sprouts and grapes. The presence of *E. coli*, *Klebsiella*, *Citrobacter*, *Edwardsiella* and *Enterobacter* may lead to diarrheal diseases. It is also a well known enteric pathogen of man in nosocomial infections. This is in conformity with the findings of Pigott¹⁸ that, the majority of human infections are caused by *Klebsiella pneumoniae* and *Klebsiella oxytoca*. The presence of *Citrobacter* spp. in the vegetables agrees with the findings of Uzeh *et al.*¹⁵ who reported *Citrobacter* spp. in carrots, cucumber, cabbage and lettuce within Lagos Metropolis. *Citrobacter* spp. is reported to be resistant to some antibiotics such as *Penicillin*. This result was in conformity with several studies conducted by many researchers in an attempt to determine or identify the microbial population responsible for vegetable spoilage. A study conducted by Manani *et al.*¹⁹ among the vegetable to identify bacteria causing spoilage, 8 samples was collected and 21 isolates were recovered from them. The bacteria found to spoil the vegetables were identified as *Klebsiella*, *Bacillus*, *E. coli*, *Staphylococcus*, *Pseudomonas* and their prevalence in vegetable sample was found to be 33.33, 23.80, 14.28, 14.28 and 14.28%, respectively. This result was also in conformity with the present research. In another study conducted by Adebayo-Tayo *et al.*²⁰ on microorganisms associated with spoilage of stored vegetables including cabbage in Uyo metropolis, Akwalbom, Nigeria showed that *Escherichia coli* (28.6%) were the most predominant bacterial isolates associated with vegetable spoilage in Uyo metropolis. This was followed by *Enterobacter* spp. (21.4%), *Staphylococcus aureus* (14.3%), *Erwinia* spp. (14.3%) and *Pseudomonas* spp. (14.3%) while *Salmonella* spp. (7.1%) was least predominant. The finding of this study was also in conformity with that of Aminu and Ali²¹ who investigated and assess the microorganisms associated with spoilage of Watermelon (*Citrillus lanatus*) who found that *Enterobacter* (23%) were the most predominant bacterial isolates associated with spoilage of water melon tested, followed by *Staphylococcus aureus* and *Salmonella* spp. (21%) each, *Escherichia coli* (19%), while *Klebsiella* spp. It was found to cause gastro-enteritis, acute gastroenteritis characterized by the symptoms of vomiting, nausea, fever, chills, abdominal pain and watery (dehydrating)

diarrhea occurring 12-24 h after ingestion of contaminated food or water²¹. Bacterial food poisoning have been implicated in the consumption of food contaminated by bacteria, most of the bacteria obtained from this study were enteric bacteria which are found in the soil and air. There is therefore the urgent need for orienting the general populace on the inherent dangers associated with consumption of these types of fruits and vegetables without thorough disinfection.

CONCLUSION

It was concluded that six bacterial species were found associated with the vegetables and fruits sold in Kaduna Central market, 2 Gram-positive (*Staphylococcus aureus* and *Streptococcus* sp.) and 4 Gram-negative (*E. coli*, *Klebsiella* sp., *Enterobacter* sp. and *Citrobacter*). These organisms have been known to produce toxins in fruits and vegetables they invade, thereby posed health hazard for their consumption. Good handling and use of clean water for washing the fruits and vegetables ought to be advocated especially by the retailers and consumers.

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