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Microbial Safety of Street Foods in Industrial Area, Nairobi

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ABSTRACT

Street foods play a significant role in feeding urban populations with cheap, accessible and nutritious foods. Most street foods vendors are not trained on food hygiene and safety. Consumption of street food can lead to food poisoning and food borne illnesses. Although studies on safety of street foods have been carried out in most developing countries, not much has been done in Nairobi Kenya. Fifty six samples classified using seven modified FAO food groups from 29 vending stalls were evaluated to investigate microbial safety of street foods. Standard microbiological methods were used for isolation, enumeration and identification of bacteria. S*almonella* was not detected in all food samples. *E. coli* was qualitatively isolated in 3 food samples from 2 locations. Expressed in $\log_{10}$ colony forming units/gram, total coliforms were detected in vegetables of all locations at unsafe levels of 4.48 and 4.3 for *Staphylococcus aureus*. Total Enterococci were 2.50 and total counts at 4.71. Meats from one location had *Staphylococcus aureus* 4.10 $\log_{10}$ CFU g^{-1} above acceptable limits of 4.00. Mixed dishes, cereals, legumes, starchy roots and beverages were safe for consumption. Using microsatellite primer confirmed each isolated group *Enterobacter aerogenes, Enterococci* species and *Staphylococcus aureus* were related. *Staphylococcal enterotoxins* d and g were in *Staphylococcus aureus* not isolated. Vegetable foods preparations require improvement as most were served uncooked as salads. The presence of microorganisms in street foods suggests hygiene practices should be improved.

Key words: Street foods, microbial safety, virulence genes, *E. aerogenes*, *Enterococci*, *S. aureus*

INTRODUCTION

FAO (2009) defines street food as food sold at various points to ease consumer access at a low cost affordable by the poor. Latham (1997) further notes that millions of people depend on a wide variety, accessible and cheap street food on daily basis. Food safety is the assurance that food will not cause harm to the consumer when prepared and consumed according to its intended use (WHO, 2001). Food safety is a progressively more essential global public health concern (WHO, 2007). WHO (2010) stated that millions of people fall sick or die as a consequence of eating unsafe food. Food borne and waterborne diarrhea diseases are principal causes of illness and globally kill an estimated 2.1 million people per year, mostly children in developing countries (WHO, 2001). WHO (2007) noted that everyone is at risk of food borne illness. Ready to eat foods and food preparation surfaces may be reservoirs for microbial contamination (Mankee *et al*., 2005; Ghosh *et al*., 2007; Christison *et al*., 2008). Various microorganisms of public health concern including faecal coliforms, *Escherichia coli*, *Staphylococcus aureus*, *Salmonella* species and

Bacillus cereus have been tested in street foods of some African countries. *Escherichia coli* and *Staphylococcus aureus* were recovered in proportions of the food, water, hands and surface swabs tested in Harare, Zimbabwe (FAO/WHO, 2005). Street foods can in addition be sources of enteropathogens (Mensah *et al*., 2002). Cooked food should contain no more than 10^6 CFU g^{-1} TVC, *E. coli* should not exceed 10 CFU g^{-1} upon analysis (KBS, 2003), 10^4 CFU g^{-1} of *Enterobacteriaceae* and greater than 10^4 CFU g^{-1} of *Staphylococcus aureus* (Gilbert *et al*., 2000; NSW Food Authority, 2009). However there are limited studies on specific hazards posed by microorganisms of public health concern in street. Pathogenicity and virulence of an organism are regulated by virulence coding genes present in the genomic regions known as pathogenicity islands (Hacker and Kaper, 2000). Contamination of food with enterotoxigenic *Staphylococcus aureus* causes Staphylococcal enterotoxins (SEs) intoxication when growth and toxin production conditions are met which is associated with symptoms like vomiting and diarrhoea. Major serological enterotoxins that have been characterized are: Staphylococcal enterotoxin a Staphylococcal enterotoxin b Staphylococcal enterotoxin c Staphylococcal enterotoxin d and Staphylococcal enterotoxin e (Robbins *et al*., 1974) and recently Staphylococcal enterotoxin g, Staphylococcal enterotoxin h, Staphylococcal enterotoxin i, Staphylococcal enterotoxin j, Staphylococcal enterotoxin k, Staphylococcal enterotoxin l, Staphylococcal enterotoxin m, Staphylococcal enterotoxin n, Staphylococcal enterotoxin o, Staphylococcal enterotoxin p, Staphylococcal enterotoxin q and Staphylococcal enterotoxin u (Letertre *et al*., 2003) studies on strain distribution are important in order to reveal influence of hygiene and diversity or commonality of contamination sources.

This study was carried out to evaluate the microbial safety of street food prepared and vended on site in the streets of Industrial area, Nairobi. No information is also available on the virulence or pathogenicity of microorganisms isolated from street food matrices. There was need to study the strain distribution and pathogenicity of presumptive food pathogens.

MATERIALS AND METHODS

Study design and study area: This was a cross sectional study carried out between March and August year 2011. The study was carried out in Industrial area, Nairobi city Kenya (Coordinates: 1°17'S 36°49'E). Nairobi is the largest city in East Africa with 3.1 million people and an annual population growth rate of 4% (KNBS, 2010). The eating places with food prepared on site in the major roads with street food in the Industrial area were selected to form the study area as follows. These included Enterprise road, Lunga lunga road, Likoni road, Nanyuki road and Ricky road.

Sampling procedure and sample size: The sampling was purposive based on availability of street vended food prepared on site in the major roads of Industrial area of Nairobi city. Seven modified FAO food groups (FAO, 2011) in Table 1 including; cereals, mixed dishes, starchy roots, beverages, vegetables, meats and legumes were sampled. Sample size was calculated as per Pfeiffer (2002) using a prevalence of 5% in microbial contamination as reported by Muganga (2001) on street food. Therefore, 56 samples were sampled and analyzed in this study. Food samples were collected in sterile polythene bags. All samples were transported to the University of Nairobi department of Food Science, nutrition and technology microbiology laboratory within three hours of collection on ice in cooler boxes.

Microbial analysis: Samples were blended by use of a stomacher (400 laboratory blender type BA 7021 England, Senard Medical London SEI IPP UK). Serial dilutions were prepared, vortexed (Genie, Bender and Hobein AG Zurich, Switzerland) and then, inoculated on selective media.

Table 1: Modified FAO grouping for food used in this study

Consumed food	Food sampled
Cereal	Ugali, chapati, rice, roast maize, mandazi
Mixed dishes	Boiled and stewed, muthokoi, githeri
Legume	Boiled and stewed beans and green grams
Vegetables	Cabbage, kales, salads/kachumbari[a]
Starchy root	Boiled, sweet potato and arrow roots, potato fries
Meat	Stewed, beef, mutton, fried fish, roast mutura[b]
Beverages	Fermented porridge, unfermented porridge, boiled milk, beef soup
Vegetables	Coriander, tomato, red onion, capsicum, red/green chillies, cabbage, kales

[a]Pepper, cabbages, capsicum, carrots, tomatoes and red onion grated, [b]Ruminants intestines casing and stuffed with blood and minced meat (FAO, 2011)

Enumeration was then done (Gerber and co. AG Zurich, Suisse) after incubation for plates with 30-300 colonies. Isolation was then done based on colony morphology followed by purification by streaking three times. Gram reactions (3% KOH), catalase reaction (3% H_2O_2; VWR International), and coagulase test for *Staphylococcus aureus* were then performed. Plate Count Agar (Oxoid Ltd., England) was used for enumeration of total plate count by pour plate method. Violent Red Bile Agar (VRBA) (Himedia Laboratories pvt, India) was used for enumeration of total coliforms (spread plate). Pinkish-red colonies were enumerated to indicate total coliforms. Verification of rod shaped by microscopy, catalase test (3% H_2O_2, VWR International), gram reactions (3% KOH, Sigma-Aldrich) tests were done to characterize *Enterobacteriaceae*. Specific coliform organisms were differentiated with IMViC tests (Harrigan and McCance, 1976). Tellurite agar was used for enumeration of *Enterococci* species (pour plate method for 42°C for 24 h). Colonies were enumerated to indicate total Enterococci after incubation for 24 h at 42°C. Baird Parker Agar (Himedia Laboratories pvt, India) was used for enumeration of *Staphylococcus aureus*. Typical colonies 2-3 mm in diameter, jet black to gray black surrounded with an opaque halo and clear zone were enumerated to indicate presumptive *Staphylococcus aureus* after incubation for 24 h at 37°C. Eosin Methyl Blue Agar Levine (Oxoid Ltd, England) was used for qualitative test of presence of *Escherichia coli*. Metallic green colonies indicated presence of *Escherichia coli*. For *Salmonella* species, pre-enrichment was done using sterile lactose broth. This was incubated at 37°C for 24 h. Ten milliliter of pre enrichment was aseptically pipetted and transferred into a jar with 100 mL sterile Tetrathionate broth (Himedia Laboratories pvt, India) and selenite cystine broth (Oxoid Ltd, England) for each and incubated at 37°C for 24 h. The resultant broth was streaked onto three *Salmonella* differential media. Brilliant green phenol lactose agar (Himedia Laboratories pvt, India), Bismuth sulphate agar (Oxoid Ltd, England) and Desoxycholate citrate agar (Oxoid Ltd, England). The typical colonies for all microbial groups were purified by streaking three times on selective media and stored 0.25 M sucrose solution at -44°C.

DNA extraction: The preserved isolates were thawed and cultured on the respective selective media. Sixty four isolates of *Enterobacteriaceae,* 33 of *Staphylococcus aureus* and 23 of *Enterococci* species were isolated and analysed. Each colony was picked and suspended in 100 µL of sterile distilled water in 1.5 mL Eppendorf tubes, heated to 95°C for 30 min and cooled immediately on ice. The solution was then be centrifuged at 15000 rpm for 5 min at 4°C in (Eppendorf centrifuge 5413 Germany). The resultant supernatant was drawn and preserved at -20°C as the template DNA.

Rep-PCR: A Rep-PCR for *Enterobacteriaceae*, *Staphylococcus aureus* and *Enterococci* species was done by the modified method described by Walczak *et al.* (2007) with primer $(GAC)_5$. The primer pre-mix which consisted 2 mM Mg^{2+} as $MgCl_2$, 0.025 μL of *Taq* polymerase, 0.2 mM dNTPs in a buffer solution, distilled nuclease free water and the DNA template were added into Eppendorf tube. This made a total reaction volume of 25 μL made in the following composition; 1 μL DNA template, 12.5 μL pre-mix, 3.25 μL GTG_5 and 8.25 μL distilled water.

***E. coli*:** *E. coli* reference strains included; STM1 (stx2), STM2 (stx1), 4115/2 (eae) and 3750/2 (stx1 and eae) pre determined to be harmful to human health as characterized by Kohler *et al.* (2008).

PCR for Staphylococcal enterotoxins: *Staphylococcus aureus* strains isolated were typed for sea, seb, sec, sed, see, seg, sei and sej. Reference strains previously isolated and characterized by Stephan *et al.* (2001) were used as for enterotoxin gene typing. These included *S. aureus* 463 (seb, seg and sei), 117 (sea, seg, sej and sei), 129 (sea, seg, sei and sej), 266 (seb, seg and sei), 216 (sec, seg and sei), 235 (sec, seg and sei), 243 (sed, seg, sei and sej) and 238 (sed, seg, sei and sej). The primers and protocols used are presented in PCR protocol for $(GAC)_5$ Table 2, PCR protocol for Staphylococcal enterotoxins in Table 3 and Sequence of oligonucleotide primers used for Staphylococcal enterotoxins and predicted lengths of PCR amplification products in Table 4.

Electrophoresis and visualization: PCR loading wells were made on 1.5% Agarose gel into which 7 μL EtBr (Carlsbad CA USA) per 100 mL agarose had been added, solidified and the gel submerged into X1 TBE buffer. A total volume of 24 μL comprising of 20 μL DNA template and 4 μL PCR loading buffer (Promega Madison USA) were loaded per well prior to electrophoresis. The separated bands were visualized with UV trans-illuminator (Vilber Laurmat-France). The visualized bands sequences were compared with those of the controls above and 1 kbp molecular marker.

Table 2: PCR protocol for $(GAC)_5$

Step	Temp. (°C)	Time	No. of cycles
Initial denaturation	94	3 min	X1
Denaturation	94	20 sec	X35
Annealing	50	1 min	
Polymerisation	72	20 sec	
Final polymerization	72	5 min	X1
Hold	4	∞	X1

All PCR reactions were done with minicycler MJ Research Inc. USA

Table 3: PCR protocol for Staphylococcal enterotoxins

Step	Temp. (°C)	Time	No. of cycles
Initial denaturation	95	5 min	X1
Denaturation	94	3 min	X35
Annealing	55	3 min	
Polymerisation	72	1 min	
Final polymerization	72	5 min	X1
Hold	4	∞	X1

All PCR reactions were done with minicycler MJ Research Inc. USA

Table 4: Sequence of oligonucleotide primers used for Staphylococcal enterotoxins and predicted lengths of PCR amplification products

Target	Primer	Sequence, 5'-3'	Product size (bp)	References
SEA	sea-1	AAAGTCCCGATCAATTTATGGCTA	219	Tsen and Chen (1992)
	sea-2	GTAATTAACCGAAGGTTCTGTAGA		
SEB	GSEBR-1	GTATGGTGGTGTAACTGAGC	164	Mehrotra *et al.* (2000)
	GSEBR-2	CCAAATAGTGACGAGTTAGG		
SEC	SEC-1	GACATAAAAGCTAGGAATTT	257	Johnson *et al.* (1991)
	SEC-2	AAATCGGATTAACATTATCC		
SED	sed-f	GTGGTGAAATAGATAGGAACTGC	385	Monday and Bohach (1999)
	sed-r	ATATGAAGGTGCTCTGTGG		
SEE	SEE-1	TAGATAAGGTTAAAACAAGC	169	Johnson *et al.* (1991)
	SEE-2	TAACTTACCGTGGACCCTTC		
SEG	SEG-1	AATTATGTGAATGCTCAACCCGATC	642	Jarraud *et al.* (1999)
	SEG-2	AAACTTATATGGAACAAAAGGTACTA		
SEI	SEI-1	CTCAAGGTGATATTGGTGTAGG	577	Jarraud *et al.* (1999)
	SEI-2	AAAAAACTTACAGGCAGTCCATCTC		
SEJ	SEJ-1	CATCAGAACTGTTGTTCCGCTAG	192	Monday and Bohach (1999)
	SEJ-2	CTGAATTTTACCATCAAAGGTAC		

Data analysis: Quantitative data on the microbial counts collected from the experiment was subjected to analysis of variance (ANOVA) using the Genstat 13th edition, VSN International. Difference among the results was compared using the Fisher's protected LSD test at 5% probability where there were significant differences amongst the means. The bands after PCR amplification and electrophoresis were compared with predetermined reference strains.

RESULTS

Microbial counts: Total coliforms counts were 4.48±0 and 3.84±0 $\log_{10}$ CFU g^{-1} in vegetable and meats foods, respectively while the lowest counts were in cereals based and legumes based foods at 2.33±0.2 $\log_{10}$ CFU g^{-1} each. *Enterococcus* species were highest in cereals based foods (2.66±0 $\log_{10}$ CFU g^{-1}) and lowest in legume foods (2.04±0 $\log_{10}$ CFU g^{-1}). The counts of *Staphylococcus aureus* were highest in vegetables (4.03±0 $\log_{10}$ CFU g^{-1}) (Table 5). The total viable microorganisms in the vegetable based foods were the highest at 4.71±0.3 $\log_{10}$ CFU g^{-1} among the seven food categories while beverages had the least counts at 3.19±0.2 $\log_{10}$ CFU g^{-1} (Table 6).

There was a significant difference in the microorganisms evaluated in vegetables from all the five locations ($p<0.05$). Ricky road had the highest mean *Staphylococcus aureus* (4.69±0.05 $\log_{10}$ CFU g^{-1}) (Table 6). Ricky, Nanyuki and Likoni roads had similar counts of *Staphylococcus aureus* while enterprise road had the least mean counts (3.13±0.15 $\log_{10}$ CFU g^{-1}). Counts of *Staphylococcus aureus* in Enterprise road were significantly similar to that in lunga lunga road. Total coliforms counts were not significantly different between Likoni and Lunga lunga road ($p<0.05$). Ricky roads had the highest coliforms count at 4.71 $\log_{10}$ CFU g^{-1} but had significantly similar levels of counts as Nanyuki road.

There was significant difference ($p<0.01$) among the microorganisms evaluated in meats from the four locations (Table 7). Enterprise road had the highest mean *Staphylococcus aureus* ($\log_{10}$ 3.67±0.05 CFU g^{-1}) while Lunga lunga road had the least mean counts (3.07±0.08 $\log_{10}$ CFU g^{-1}) in meat. Counts of *Staphylococcus aureus* and total coliforms in meat were significantly different among all the locations ($p<0.05$). Nanyuki road had the highest coliforms contamination at 4.10±0.02 $\log_{10}$ CFU g^{-1}. TVC levels in meat were significantly similar in Likoni, Nanyuki and Lunga lunga roads and for Enterprise road, Nanyuki road and Ricky road.

Table 5: Prevalence and viable counts of street foods in Industrial area, Nairobi

		Food group						
Bacteria group	Counts	Cereals (n = 12)	Mixed (n = 0)	Legumes (n = 7)	Vegetables (n = 11)	Starchy (n = 5)	Meats (n = 9)	Beverages (n = 6)
Total viable	Positive samples	5	4	7	11	5	8	5
	Log_{10} CFU g^{-1}	4.33	4.14±0.2	3.79	4.71±0.3	3.98	4.21	3.19±0.2
Total	Positive samples	1	4	2	7	2	4	0
	Log_{10} CFU g^{-1}	2.33±0	2.72±0	2.33±0	4.48±0	2.42±0	3.84±0	*
Enterococci	Positive samples	4	0	1	2	1	2	0
	Log_{10} CFU g^{-1}	2.66±0	*	2.04±0	2.50±0	2.44±0	2.40±0	*
Staphylococcus aureus	Positive samples	1	1	1	6	0	6	0
	Log_{10} CFU g^{-1}	3.27±0	3.32±0	3.37±0	4.03±0	*	3.45±0	*

*Not enumerated/not tested

Table 6: Microbial counts of street food vegetables in five locations sampled in Industrial area of Nairobi city

	Location					
Microorganism	Enterprise	Likoni	Lunga lunga	Nanyuki	Ricky	Mean
Staphylococcus	3.13±0.15^{a}	4.51±0.06^{b}	3.33±0.05^{a}	4.45±0.05^{b}	4.69±0.05^{b}	4.02
Total coliforms	4.34±0.10^{a}	4.53±0.04^{b}	4.49±0.08^{b}	4.65±0.02^{c}	4.71±0.03^{c}	4.54
Total *Enterococci*	2.04±0.09^{a}	2.72±0.04^{b}	2.04±0.14^{c}	3.44±0.06^{d}	2.27±0.02^{e}	2.67
Total viable counts	4.61±0.04^{a}	4.83±0.01^{b}	4.75±0.07^{c}	4.46±0.02^{d}	5.03±0.03^{e}	4.74

Values (log_{10} CFU mL^{-1}) with same superscript in each row are not significantly different at p>0.05

Table 7: Microbial counts in street food meats in four locations sampled in Industrial area of Nairobi city

	Location				
Microorganism	Enterprise	Lunga lunga	Nanyuki	Ricky	Mean
Staphylococcus aureus	3.67±0.05^{a}	3.07±0.08^{b}	3.22±0.03^{c}	3.33±0.06^{d}	3.30
Total coliforms	3.00±0.02^{a}	3.96±0.03^{b}	4.10±0.02^{c}	3.80±0.02^{d}	3.72
Total viable counts	3.85±0.13^{b}	3.55±0.17^{a}	3.78±0.13^{a,b}	3.77±0.14^{ab}	3.73

Values (log_{10} CFU mL^{-1}) with same superscript are similar

Escherichia coli was detected in two food groups from two locations which were mixed dish collected from Lunga lunga road and two vegetable samples from Lunga lunga and Nanyuki roads. Specific microorganisms isolated include; sixty isolates of *Enterobacter aerogenes,* 22 of *Enterococci* species and 33 of *Staphylococcus aureus*. *Salmonella* species were not detected in any of the food groups in all locations sampled in this study.

Strain distribution of microorganisms: The 60 isolates of *Enterobacter aerogenes* were analysed by a rep PCR. The isolates included 17 from vegetables, 7 from cereals, 18 from mixed dishes, 4 from starchy roots, 12 from meats and 2 from legumes. All samples had a similar single band amplified at the same electrophoretic length (Fig. 1a). A total of 22 *Enterococci* species included 13 from vegetables, 3 from cereals, 3 from meats, 2 from starchy roots and 1 from legumes. All samples had same level of single bands amplified (Fig. 1b). A total of 33 isolates of *Staphylococcus aureus* were obtained from street foods including 17 from vegetables, 2 from cereals, 10 from meats, 2 from starchy roots, 1 from legumes and 1 from mixed dishes. All samples had a similar single band

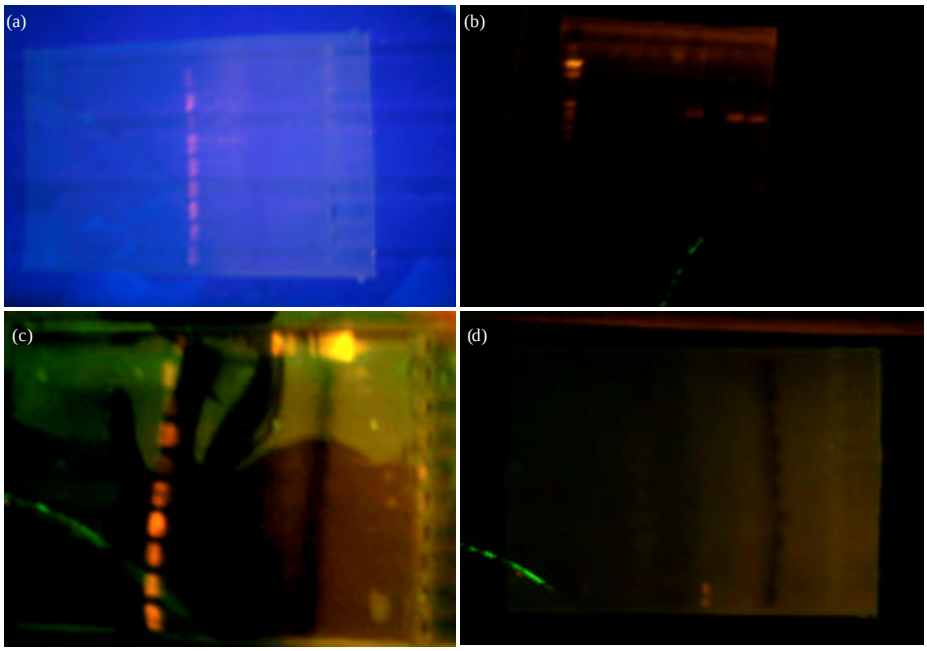

Fig. 1 (a-d): Rep-PCR gel pictures of (a) *Enterobacter aerogenes*, (b) Enterococci species (c) *Staphylococcus aureus* and (d) Reference *Staphylococcus aureus* strains

amplified at the same electrophoretic length (Fig. 1c). Figure 1d shows an amplified band of a reference strain of *Staphylococcus aureus* predetermined to process the gene coding for staphylococcal enterotoxin d.

Presence of Staphylococcal enterotoxin genes: The seven categories of foods collected from streets of industrial area of Nairobi were evaluated for the possession of staphylococcal enterotoxins. All the isolates were negative for the sed and seg. Rep PCR for other SEs did not produce amplicons. Successful amplification occurred for sed and seg.

DISCUSSION

The highest total viable counts were noted in vegetable foods at 4.71±0.3 $\log_{10}$ CFU g^{-1} among the seven food categories. This may be attributed to the fact that most vegetables were served raw in salads or only partly cooked. Beverages had the least mean bacterial counts of 3.19±0.2 $\log_{10}$ CFU g^{-1}. This could be explained by the fact that all the beverages are subjected to heat treatment during their preparation and are served hot. Cereals, mixed dishes, legumes, starchy roots and meats had varying levels of counts TVC while the highest counts was in Ricky road vegetables at 5.03±0.03 $\log_{10}$ CFU g^{-1}. Any cooked food should contain no more than 6.00 $\log_{10}$ CFU g^{-1} viable counts per gram upon analysis (KBS, 2003; Gilbert *et al*., 2000). Presence of *S. aureus*, an enterotoxin producer cause serious gastroenteritis (Balaban and Rasooly, 2000). All vegetables had levels of coliforms and *Staphylococcus aureus* at the unacceptable levels above 4.00 $\log_{10}$ CFU g^{-1}. Meat (fish) from Ricky road had levels of *Staphylococcus aureus* at 4.10 $\log_{10}$ CFU g^{-1} slightly above the acceptability threshold of 4.00 $\log_{10}$ CFU g^{-1} (KBS, 2003; Gilbert *et al*., 2000). In terms of contamination with *Enterococcus* species, foods were safe at levels below 4.00 $\log_{10}$ CFU g^{-1} with the highest contamination being

noted in Nanyuki road at 3.44±0.06 $\log_{10}$ CFU g^{-1} with other foods ranging 2.04-2.66 $\log_{10}$ CFU g^{-1}. Enterococci species were not isolated in mixed dishes and beverages. The levels enumerated may indicate a possible contamination during handling after cooking. The counts in vegetables at 4.03 $\log_{10}$ CFU g^{-1} and meats at 3.45 $\log_{10}$ CFU g^{-1} suggest a contamination probably before serving.

Ricky road was noted to consistently have the highest count levels in vegetables with *Staphylococcus aureus* at 4.69 $\log_{10}$ CFU g^{-1}, total coliforms at 4.71 $\log_{10}$ CFU g^{-1} and total viable counts at 5.03 $\log_{10}$ CFU g^{-1} in vegetables which could be attributed to practices of food handling and the initial contamination of the raw material sourced in that area. *Staphylococcus aureus* suggests a contamination which emanates from food handling which might have occurred in the street foods during handling, processing or vending. *Staphylococcus aureus* being part of the micro flora present on/in several parts of the human body is a good indicator of contamination due to poor personnel hygiene practices (Nester *et al*., 2001). Fish from Nanyuki road were not acceptable for consumption as they had 4.10 $\log_{10}$ CFU g^{-1} of *Staphylococcus aureus* while those from Enterprise, Lunga lunga and Ricky roads are safe for consumption (KBS, 2003; Gilbert *et al*., 2000).

Mensah *et al*. (2002), in Ghana found that foods particularly heavily contaminated with *Staphylococcus aureus* since they are handled excessively after cooking.

Total coliform counts at 4.48 $\log_{10}$ CFU g^{-1} were high in vegetables which may be attributed to the fact that most vegetables were served raw as uncooked salads. Similar results were reported in a study carried out by Amponsah-Doku *et al*. (2010) in raw lettuce with range (4.35-9.20 $\log_{10}$ CFU g^{-1}) vended in the streets of Ghana. *Enterobacteriaceae* are useful indicators of hygiene and post processing contamination of heat-processed foods. Their presence in high numbers (>10^4 g^{-1}) in ready to eat foods indicates that either contamination or under processing. The level of bacterial loading on lettuce and other raw vegetables may originate from the farm irrigation water (Mensah *et al*., 2001; Obiri-Danso *et al*., 2005). The contamination could also be attributed to sub standard cutting and preparation practices, particularly poor hygienic conditions, of the premises that may result from, rubbish, sewage and other noxious substances present in the vicinity (WHO, 2007). Bhaskar *et al*. (2004) and Mosupye and von Holy (2000) had observed that bacteria from dirty dish washing waters and other sources can adhere to utensil surfaces and constitute a risk for contamination during food vending.

The coliforms counts were at unacceptable levels in ready to eat foods as enumerated in vegetables all >4.00 $\log_{10}$ CFU g^{-1}. Only one food (rice) was detected to be contaminated with coliforms (<2.00 $\log_{10}$ CFU g^{-1}). Contamination of mixed dishes and legumes with coliforms albeit at low levels could have emanated from post cooking handling. The counts of coliforms at unacceptable levels may be attributed to inadequate handling: Where food like fried fish as in this case is displayed and sold in the open air and handled by vendors with bare hands. This agrees with Tambekar *et al*. (2009) who found severe contamination of displayed food through handling. Presence of coliforms in street foods might also be due to water used for cooking and serving which was contaminated with faecal coliforms as found by Khalil *et al*. (1994).

The mixed dish collected from Lunga lunga road (1/56) and vegetables in both Lunga lunga and Nanyuki roads (2/56) tested positive for *Escherichia coli*. This consisted of 5.3% of all the samples and is slightly higher compared to a study that reported in 3% of Korean street foods (Cho *et al*., 2010). This is in contrast with a study conducted in the street food of cape coast Ghana by Annan-Prah *et al*. (2011) where all sampled food tested positive for *Escherichia coli*. *Escherichia coli* have been previously detected in 48% of foods sampled from Korogocho slums of

Nairobi (Muganga, 2001). *Salmonella* species *were* not detected in any of the food consumption food group across all roads sampled in this study. The consumption of street food cannot be stopped on hygienic grounds (Tambekar *et al.*, 2009). These practices could be improved to ensure more safety. Contamination is mainly due to poor quality of water used for dilution and prevailing unhygienic condition related to improper washing of fruits, vegetables and utensils, inadequate storage in unhygienic places and personal hygiene by vendors (Das *et al.*, 2010).

This study demonstrates that the foods were safe for human consumption which can be attributed to the fact that the foods were mostly served hot and only held for 1-3 h before serving. Presence of coliforms and *Staphylococcus aureus* indicates a possibility of secondary contamination. The study by Muganga (2001) indicated that majority of highly contaminated foods were prepared on site where holding time was 1.5-12 h. *Bacillus cereus*, *Staphylococcus aureus*, or *Enterobacteriaceae* have been reported not to survive in breakfast, snack foods and porridge prepared and sold within 2-3 h at 50-90°C, a temperature range over which most vegetative bacteria (Mensah *et al.*, 2002). Such long time heat treated foods have acceptable levels of microbial contamination. Annan-Prah *et al.* (2011) in Ghana also found kebabs, fried fish and beans with gari to had acceptable levels of microbial contamination provided that they are not excessively and poorly handled after cooking (Yeboah-Manu *et al.*, 2010). Von Holy and Makhoane (2006) also found that the quality of street foods in South Africa is not as bad as previously conceived.

According to the reviewed literature, this is the first study evaluating the strain distribution of microorganisms isolated from street food. From the PCR analysis, the isolates from each of *Staphylococcus aureus*, *Enterococci* species and *Enterobacter aerogenes* from street food were similar strains as shown by rep PCR with primer GTG_5. The similar bands after from PCR analysis indicate that all isolates were similar strains and could have originated from a similar source; processing water, raw material and cooked food cross contamination or contamination from the body of food handlers' into the food.

This is also the first study to evaluate the pathogenicity of *Staphylococcus aureus* strains in street food in Kenya. However, none of the isolates from the street food was seen to possess genes coding for production of staphylococcal enterotoxins *sed* and *seg*. The absence of the genes coding for production of the above staphylococcal enterotoxins implies the *S. aureus* isolated from street foods are not a likely threat to human health. The presence of the microorganisms is an indicator of poor hygiene and not necessarily a threat to human health. More studies are required to evaluate the safety of water used in the preparation of street foods of Nairobi and the evaluation of presence of other enterotoxins in the street food.

CONCLUSION

Most of the foods sampled and analyzed in this study including cereals, legumes, starchy roots, mixed dishes, legumes and meats from Enterprise, Ricky, Lunga lunga and Likoni roads met microbiological standards of ready to eat foods. The presence of unacceptable levels of coliforms and *Staphylococcus aureus* in the vegetables and *S. aureus* in meat (fish) from Nanyuki road suggest hygiene practices need be improved to assure food safety upon consumption. The vendors prepared and served the food which is mostly preferred hot and this factor combined with preparation on site could result in the safe street food as revealed by this study. *Staphylococcus aureus* isolated from street food do not possess enterotoxigenic genes that code for production of sed and seg. Street foods in Nairobi are therefore not likely to pose a public health risk through *S. aureus* enterotoxigenic food borne illness. The isolated microbial strains were found to be similar at molecular level,

suggesting a similar source of contamination. This indicates the need to improve hygiene practices in the preparation and handling of street foods in order to avoid contamination resulting from this similar source.

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