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Evaluation of microbial contamination of consumed fruits and vegetables salad (*Kachumbari*) around Egerton University, Kenya

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ABSTRACT

With increased student population and health consciousness in and around Egerton University, Kenya, there has been an upsurge in the vending of fruits and vegetable salad (*Kachumbari*). However, there have been safety concerns, and increased infections have been reported. This study evaluated the microbiological quality of salads served around Egerton University, particularly targeting *Escherichia coli* and *Salmonella* spp. contamination. Thirty salad samples, five each from different sampling zones (Egerton University gate, Nakuru bus stage, Nakuru town hawkers, food kiosks around Egerton University, Njokerio vendors, and Njoro town vendors), were collected randomly. The *E. coli* was determined on MacConkey Agar and Brilliant Green Bile Broth, while its confirmation was done on Eosin Methylene Blue agar. *Salmonella* spp. was determined on Salmonella Shigella Agar and confirmed using Triple Sugar Agar slants. Approximately 80% of the samples tested positive for *E. coli* with a mean value of $\log_{10}$ of 3.047 colony forming units (CFU)/g. On the other hand, 70% of the samples tested positive for *Salmonella* with a mean of $\log_{10}$ 2.067 CFU/g. These values were above the Kenya Bureau of Standards of 10 CFU/g for *E. coli* and absence of *Salmonella* in fruits and vegetable salads. The present study revealed the potential hazards of street-vended salad and the illnesses affecting consumers may be attributed to these street-vended foods. Therefore, a Hazard Analysis and Critical Control Points Principles study is essential to identify the source of microbial contamination in the salads to guide the implementation of food safety measures by the public health officials.

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1. Introduction

Street-vended food including ready-to-eat food and beverages, which are prepared and sold outside established buildings or in makeshift structures by street vendors are gaining importance in Kenya due to their economic contribution in job creation and reducing food insecurity (1). In particular, salads, which can be defined as a food made primarily of a mixture of raw vegetables and/or fruits are gaining prominence due to their health and nutritional benefits. However, safety and quality are the two common concerns with regard to street-vended foods (2,3). Poor hygiene and sanitation practices are one of the major bottlenecks in street food vending. These predispose consumers to foodborne diseases, particularly where the products are consumed without further processing, washing or peeling (3,4). The water used for washing and rinsing the fruits and vegetables is of questionable quality and could serve as a source of contamination and cases of fruits and vegetable contamination with microorganisms during production; harvesting, packing and distribution have been reported. For instance, *Salmonella* has been found in a wide range of "organically grown" products including beans, peas, sunflower seeds, and alfalfa (5). In the USA, 2 out of 111 lettuce analyzed were found to contain *Salmonella*, and 3 out of 66 green onion samples contained *Shigella* (6). Moreover, microorganisms

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multiply faster on cut produce, owing to the greater availability of nutrients and water.

Street food vendors do use gloves in handling food in attempts to reduce the transmission of foodborne pathogens. However, most gloves are permeable to bacteria during actual use, leading to the presence of the microbial contaminants in food (7). Therefore, continued surveillance of street-vended food is of importance to assure consumer safety, irrespective of the incorporation of handling methods intended to reduce food contamination.

Around Egerton University, Kenya, with recent increases in student population, there has been a steady rise in the number of street food vendors selling to University students and staff and the local community. All around Kenya, consumption of street-vended food has been implicated as a vehicle for transmission of infectious microorganisms in foodborne outbreaks, including areas around Egerton University (1,2,3). The present study was conducted to evaluate the microbiological quality of salads sold near Egerton University, Kenya, targeting the detection of *Escherichia coli* and *Salmonella* spp. and the results are here reported.

2. Materials and methods

The study was conducted at Egerton University, Nakuru County in Kenya. Random sampling procedure was adopted to select six zones (Egerton University gate, Nakuru bus stage, Nakuru town hawkers, and food kiosks around Egerton University, Njokerio vendors, and Njoro town vendors). The samples taken were obtained using proportionate representation according to size.

First, observations on *kachumbari* preparation were done. Then, a total of 30 samples were collected from six different zones of the areas around Egerton University, Kenya. Samples (100 g of *kachumbari*) were transported to the laboratory in sterile 500 ml sample bottles under aseptic conditions and were kept in an icebox maintained at 6-8 °C. For the analysis, 25 g of each salad sample was weighed and transferred to 225 ml of sterile buffered peptone water then blended. Serial dilutions were conducted, and aliquots of buffered peptone water were then inoculated onto the respective media then incubated.

For the cultivation of cells, 1 ml of the homogenate was pipetted and dispensed in 9 ml of sterile peptone water. Serial dilutions were similarly carried out up to 10^{-4}. Subsequently, a 0.1 ml aliquot was then drawn and dispensed into a sterile Petri dish and pour plated with violet red bile agar (Oxoid Ltd, England). Distinct red colonies were picked with a wire loop and streaked onto eosin methylene blue Agar (Oxoid Ltd, England). Colonies with metallic green sheen were recorded as *E. coli*.

For the enumeration of *Salmonella*, 25 g of the sample was inoculated into 225 ml of peptone water as pre-enrichment and incubated at 37 °C for 24 hours. Consequently, 1 ml was then pipetted and transferred into 10 ml of Rappaport Vassiliadis medium (Oxoid Ltd, England) and incubated at 42 °C for 24 hours. A loopful of the growth was then streaked onto both xylose lysine deoxycholate (XLD) and Brilliant Green Agar (BGA) (Oxoid Ltd, England). The agar plates were incubated at 35 °C for 24-48 hours. The colonies that appeared on XLD and BGA were then streaked onto MacConkey agar (Oxoid Ltd., England) and the purified colonies inoculated on Triple sugar iron agar (Oxoid Ltd., England) slants by stabbing the butt and streaking the slant.

Data analysis involved descriptive analysis (mean, minimum, and maximum) for each category of the group. Microbiological counts obtained from *E. coli* and *Salmonella* counts were converted to base-10 logarithm of colony forming units (CFU) per gram of each sample.

3. Results

The *kachumbari* preparation flow diagram is presented in figure 1. The process involved washing of the fruits and vegetables with water whose quality could not be ascertained. In the case of onions, after peeling the outer skin, they were chopped into small pieces then mixed to make *kachumbari* without washing them. The cleanliness of the vendors was not proper, and their health status could not be established hence they could potentially serve as sources of contamination. The handling of the *kachumbari* by most vendors was also not proper as it was neither covered nor refrigerated. Therefore, dust particles could fall on the foods and serve as sources of contamination and the contaminants would grow amply at ambient temperatures.

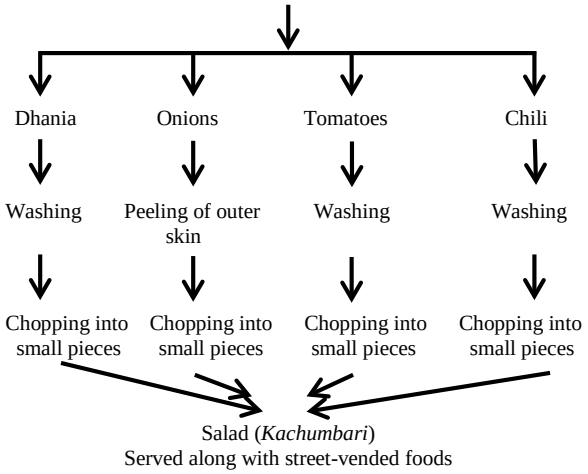

Figure 1. Flow diagram for the preparation of salad (*kachumbari*) sold around Egerton University, Kenya

Table 1. Mean concentration ranges of *E. coli* spp. and *Salmonella* spp. in *kachumbari* samples around Egerton University, Kenya

Sampling zone/microbial count	*Salmonella* spp. counts Log_{10} CFU/g	*E. coli* spp. counts Log_{10} CFU/g
Egerton University gate	1.016 ± 0.486	2.782 ± 0.240
Nakuru town bus stage	2.727 ± 0.432	3.146 ± 0.182
Nakuru town hawkers	2.596 ± 0.470	3.146 ± 0.220
Food kiosks around Egerton University	0	2.676 ± 0.173
Njokerio vendors	1.639 ± 0.315	3.051 ± 0.221
Njoro town vendors	2.359 ± 0.321	3.242 ± 0.238

Results are means ± standard deviations of triplicate samples. CFU: Colony forming units, *E. coli: Escherichia coli*

3.1. Raw dhania, onions, tomatoes and chili

The microbiological analysis of the salad samples indicated that *kachumbari* was contaminated with pathogens. The *E. coli* was present in 80% of the samples analyzed and in all the sampling zones while *Salmonella* tested positive in 70% of the samples and was prevalent in five of the six sampling zones (Table 1). The *E. coli* concentrations in the samples ranged between log_{10} of 2.676 CFU/g in food kiosks and log_{10} of 3.242 CFU/g in samples that were collected from Njoro town vendors (Table 1). The mean average for *E. coli* was log_{10} of 3.047 CFU/g, which was several orders high compared to the Kenya Bureau Standards recommended value of < 10 CFU/g. The concentrations of *Salmonella* ranged between log_{10} 1.016 CFU/g and log_{10} 2.727 CFU/g with an average of log_{10} 2.067 CFU/g. *Salmonella* should be negative in food samples, and for the concentrations detected in this study, it is an indication of a health hazard that needs to be addressed. Whereas *E. coli* was detected in all the sampling zones, *Salmonella* was not detected in the food kiosks (Table 1). The food kiosks also had lower levels of *Salmonella*, indicating that coupled with hygiene, food safety can be maintained if food could be sold in kiosks compared to open places.

4. Discussion

The present study aimed at assessing the prevalence of *Salmonella* spp. and *E. coli* in the most consumed salad (*kachumbari*) around Egerton University, Kenya. The result showed that in the 30 samples examined; *Salmonella* spp. was present in 21 samples while *E. coli* was present in 24 samples. This result showed that *E. coli* was more predominant than *Salmonella* spp. The presence of these pathogens in the salads could be a direct reflection of sanitary quality of the cultivation water, harvesting, transportation, storage, and processing of the salad (8). Freshly consumed vegetables especially those used in salad mixtures, have been implicated in food poisoning and thus; hazardous to the health of the consumers. This could be linked to the fact that most of these vegetables are consumed without being subjected to thermal process or even thorough washing (3,9). *Salmonella* spp. and *Shigella* spp. are non-lactose fermenters usually associated with water contamination. Contamination with these organisms could arise from washing vegetables with contaminated water or handling of the vegetables by infected workers (3,4). According to a study by Amponsah-Doku et al. (10) on the bacterial contamination of lettuce (ingredient in the salad) and at production sites, markets and street food restaurants in the city of Kumasi, and Ghana in general, the levels of thermotolerant coliforms on lettuce was increased by 18.0%, while *Enterococci* numbers reduced by 64.0% from the farms to the street foods. This indicates that contamination of *kachumbari* could have occurred during transporting, washing, and processing of the vegetables before consumption.

Vegetables are commonly associated with food poisoning, and they harbor disease-causing organisms. Processing of raw vegetables into salads for sale creates conducive environments and opportunities for the multiplication of pathogenic microorganisms on the salads (3,11). This is because the salads still retain enough moisture to promote microbial growth, and also the natural protective covering on the leaves against the entry of microorganisms may have been lost during harvesting, storage, transport, and processing (3,4). The *kachumbari* may also have undergone some fermentation during sale and the increased acidity could promote the growth of certain microbes such as *Salmonella* spp. which grow well in optimal pH of 4.2-8.2 (11).

Vegetables can be contaminated with pathogens from animal and human reservoirs and the environment as a result of production practices. A major source of contamination is organic fertilizer (e.g. manure and municipal sludge) and fecal contaminated water (7). The need for microbial assessment of these vegetables for production of food salads and for other use cannot be over emphasized to reduce possible contamination (12). *Kachumbari* handled in buildings (kiosks) where they are not exposed to sunlight, and dust particles had less contamination, indicating the need to handle food in kiosks or in buildings.

5. Conclusion

The high bacterial load and presence of these organisms especially *E. coli* and *Salmonella* in the

kachumbari could serve as an indicator for the need to promote awareness about the possible health hazards that could be due to poor handling of these vegetables. There is, therefore, the need for regulatory bodies to ensure that microbiological standards are established and practiced by farmers and marketers for the handling and distribution of salads. Despite the relatively small sample size of this survey, the results obtained have demonstrated that the microbiological quality of fresh-cut leafy salad vegetables and fruits sold in Egerton and its surrounding is not impressive. However, to ensure the continued safety of these products into the future, it is critical that food safety control measures are effectively implemented, and a Hazard Analysis and Critical Control Points Principles study is essential to identify contamination sources.

Conflict of Interests

Authors have no conflict of interest.

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References

1. Gitahi MG, Wangoh J, Njage PMK. Microbial safety of street foods in industrial area, Nairobi. Res J Microbiol 2012; 7: 297-308.
2. World Health Organization. The five keys to safer food. Available at: http://www.who.int/foodsafety/about/Flyer_keys.pdf. Cited Mar, 18 2016.
3. Muhonja F, Kimathi GK. Assessment of hygienic and food handling practices among street food vendors in Nakuru Town in Kenya. Science Journal of Public Health 2014; 2: 554-559.
4. Rane S. Street vended food in developing world: Hazard analyses. Indian J Microbiol 2011; 51: 100-106.
5. Andrews WH, Wilson CR, Poelma PL, et al. Bacteriological survey of sixty health foods. Appl Environ Microbiol 1979; 37: 559-566.
6. US Food and Drug Administration (FDA). FDA Survey of imported fresh produce - Import produce assignment (DFP # 05-16). Available at: https://www.fda.gov/Food/GuidanceRegulation/GuidanceDocumentsRegulatoryInformation/ProducePlantProducts/ucm118884.htm. Cited Aug, 10 2015.
7. Montville R, Chen Y, Schaffner DW. Glove barriers to bacterial cross-contamination between hands to food. J Food Prot 2001; 64: 845-89.
8. Beuchat LR. Pathogenic microorganisms associated with fresh produce. J Food Prot 1996; 59: 204-216.
9. Lund BM. Ecosystems in vegetable foods. J Appl Microbiol 1992; 73: 115s-126s.
10. Amponsah-Doku F, Obiri-Danso K, Abaidoo RC, et al. Bacterial contamination of lettuce and associated risk factors at production sites, markets and street food restaurants in urban and peri-urban Kumasi, Ghana. Sci Res Essays 2017; 5: 217-223.
11. Ameko E, Achio S, Alhassan S, et al. Microbial safety of raw mixed-vegetable salad sold as an accompaniment to street vended cooked rice in Accra, Ghana. Afr J Biotechnol 2012; 11: 11078-11085.
12. Nma ON, Oruese OM. Bacteriological quality of street-vended Ready-to-eat fresh salad vegetables sold in Port Harcourt Metropolis, Nigeria. Academia Arena 2013; 5: 65-75.