

Journal of Food Quality and Hazards Control 7 (2020) 18-26

Microbial Quality of Ready-to-Eat Street Vended Food Groups Sold in the Johannesburg Metropolis, South Africa

C.V. Asiegbu, S.L. Lebelo, F.T. Tabit ^{*}⊠[™]

Department of Life and Consumer Sciences, University of South Africa, Cnr Christiaan de Wet Road and Pioneer Avenue, Florida, Roodepoort 1710, Johannesburg, South Africa

HIGHLIGHTS

- Out of 205 ready-to-eat Street-Vended Foods (SVFs) samples, 85.37% had aerobic growth.
- The dominant genus in ready to eat SVFs was *Pseudomonas* spp., followed by *Escherichia* spp. and *Bacillus* spp.
- Prevalence rates of *Listeria monocytogenes*, *Staphylococcus aureus*, *Salmonella* spp., and *Escherichia coli* O15:H7 were 46.36, 31.8, 21.8, and 1.8%, respectively.
- The microbial safety of ready-to-eat SVFs sold in the Johannesburg Metropolis remains a serious public health concern.

Article type	
Original article	

Keywords Bacterial Load Polymerase Chain Reaction Fast Foods Food Safety South Africa

Article history Received: 6 Mar 2019 Revised: 27 Jul 2019 Accepted: 8 Aug 2019

Acronyms and abbreviations CFU=Colony Forming Unit PCR=Polymerase Chain Reaction SVF=Street Vended Food

ABSTRACT

Background: In many developing countries, the risk of contracting a food-borne disease is high after consuming contaminated ready-to-eat Street-Vended Foods (SVFs). The main objective of this research was to assess the microbiological quality of SVF groups sold in the Johannesburg Metropolis, South Africa.

Methods: A stratified random sampling procedure was used for collecting the ready-toeat SVF samples. Methods prescribed by the International Organization for Standardization (ISO) were used for analyses for aerobic colony count, Enterobacteriaceae count, presence of *Escherichia coli* O15:H7, detection of *Salmonella*, *Staphylococcus aureus*, and *Listeria monocytogenes*. The bacterial isolates were identified by 16S rRNA gene sequencing. Data analysis was done using IBM SPSS Statistics V25.0.

Results: Of the 205 ready-to-eat SVF samples, 85.37% had aerobic growth. The vast majority (78.18%) of the 110 ready-to-eat SVF samples had Enterobacteriaceae growth. From the 110 SVF samples, the prevalence rates of *L. monocytogenes*, *S. aureus*, *Salmonella* spp., and *E. coli* O15:H7 were 46.36, 31.8, 21.8, and 1.8%, respectively. There was no statistical significant difference (*p*>0.05) in the prevalence rates of *L. monocytogenes*, *S. aureus*, *Salmonella* spp., and *E. coli* O15:H7 in the various SVF groups.

Conclusion: Based on the findings of this study, the microbial quality and safety of ready-to-eat SVFs sold in the Johannesburg Metropolis remain a serious public health concern. Hence, it is necessary to educate street food vendors and enforce food safety legislation in the street food sector in the country.

© 2020, Shahid Sadoughi University of Medical Sciences. This is an open access article under the Creative Commons Attribution 4.0 International License.

common (Asiegbu et al., 2016; Oguttu et al., 2014). Gen-

erally, the ready-to-eat SVF sector plays a significant role

Introduction

In many developing countries, the sale and consumption of ready-to-eat Street-Vended Foods (SVFs) is

* Corresponding author (F.T. Tabit) [™] E-mail: tabitft@unisa.ac.za OPCU UD: http://doi.org/0000.0001.80

ORCID ID: https://orcid.org/0000-0001-8004-8415

To cite: Asiegbu C.V., Lebelo S.L., Tabit F.T. (2020). Microbial quality of ready-to-eat street vended food groups sold in the Johannesburg Metropolis, South Africa. *Journal of Food Quality and Hazards Control.* 7: 18-26.

in granting low-income population groups access to cheap food. Consumers greatly appreciate such outlets for their convenience as well as the unique taste (Kibret and Tadesse, 2013).

Although, the majority of SVF consumers seem to have little confidence in the safety of the offerings, this issue does not affect their preference for these foods (Asiegbu et al., 2016). Unfortunately, the risk of contracting foodborne diseases due to the consumption of SVFs harboring microbial contaminants is relatively high and poses a serious public health risk (Alimi, 2016; Campos et al., 2015; Kothe et al., 2016).

Food-borne pathogens could grow rapidly in SVFs and may cause diseases in consumers upon consumption (Rane, 2011). For instance, a listeriosis outbreak was recorded in South Africa from January 2017 to March 2018 in which 674 cases were recorded with 183 deaths (NICD, 2018; WHO, 2018). Despite the health risks posed by the consumption of SVFs, the number of vendors selling foods at street corners has continued to increase over the years, due to the urbanization of rural communities, rural-to-urban migration, urban population growth, and changing lifestyles (Khongtong et al., 2014).

Currently, the production, sale, and consumption of SVFs continue to flourish in many neighborhoods in the Johannesburg Metropolis (Asiegbu et al., 2016). The principle objective of the research was to assess the microbiological quality of selected ready-to-eat SVF groups sold in various neighborhoods in Johannesburg Metropolis.

Materials and methods

Samples

A cross-sectional study was conducted from which a stratified random sampling procedure was used to gather SVFs from street food vending sites in the Johannesburg Metropolis, South Africa. Ready-to-eat SVF samples (n=205) of different food groups (Table 1) were purchased from randomly selected street food vendors in each stratum over a period from February 2016 to August 2017. In total, 205 samples were used for aerobic colony counts, and 110 samples were used for Enterobacteriaceae and pathogen detection analyses. Street food samples used for aerobic colony counts and subsequently became unavailable as the study progressed, were excluded from the Enterobacteriaceae and pathogen detection analyses.

Enterobacteriaceae and aerobic mesophilic counts

Each sample was analyzed for aerobic colony count (ISO, 2013) and Enterobacteriaceae count (ISO, 2004).

For the aerobic colony count and Enterobacteriaceae counts, 25 g of each food sample was mixed with 225 ml of sterile buffered peptone water (Biolab, South Africa) and homogenized for 2 min in a stomacher at medium speed. One ml of the serial dilutions (10⁻¹ to 10⁻⁵) of each sample was plated on plate count agar (Merck, South Africa), and incubated at 37 °C for 24 h. Plates containing 30-300 colonies were counted to obtain aerobic colony count as Colony Forming Unit (CFU). Similarly, one ml of the serial dilutions of each sample was plated on Violet Red Bile Dextrose Agar (Merck, South Africa), and incubated at 37 °C for 24 h. Plates containing 30-300 colonies were also counted to obtain the Enterobacteriaceae counts.

Detection of Escherichia coli O15:H7

According to ISO (2001), enrichment for *E. coli* O15:H7 was conducted by mixing 25 g of each food sample with 225 ml of *E. coli* HiCromeTM enrichment broth (Sigma-Aldrich, Johannesburg South Africa), homogenized and incubated at 37 °C for 21 h. Thereafter, 200 µl of each mixture was plated on prepared RAPID' *E. coli* 2 agar plates (Biolab, South Africa) containing 0.01 g/L pimaricin (Promepharma, Johannesburg, South Africa) and incubated at 37 °C for 24 h. Plates showing pink to violet colonies were recorded as positive plates. The *E. coli* Latex Test kit (Hampshire, United Kingdom) was used to confirm the identities of presumptive colonies.

Detection of Salmonella spp.

In order to detection of *Salmonella* spp., enrichment step was conducted by mixing 25 g of each food sample with 225 ml of *Salmonella* enrichment broth (Sigma-Aldrich, Johannesburg, South Africa), homogenized, and incubated at 37 °C for 21 h (ISO, 2002). Thereafter, 200 µl of each mixture was plated on *Salmonella* Chromogen Agar (Sigma-Aldrich, Johannesburg, South Africa) plates containing 0.01 g/L pimaricin (Promepharma, South Africa) and incubated at 37 °C for 24 h. Plates showing magenta colonies were recorded as positive plates. The identities of colonies were confirmed by means of a latex agglutination test using the Oxoid Salmonella Latex Test (ThermoFisher, Hampshire, United Kingdom).

Detection of Staphylococcus aureus

Enrichment for *S. aureus* was carried out by mixing 25 g of each food sample with 225 ml of modified Giolitti and Cantoni broth (Sigma-Aldrich, Johannesburg, South Africa), homogenized, and incubated at 37 °C for 21 h (ISO, 2003). Thereafter, 200 µl of each mixture was plated on Baird Parker Agar (ThermoFisher, Hampshire,

United Kingdom) plates containing 0.01 g/L pimaricin (Promepharma, Johannesburg, South Africa) and incubated at 37 °C for 24 h. Plates showing grey-black shiny colonies with an opaque zone around each colony were recorded as positive plates. The identities of colonies were confirmed by means of a latex agglutination test using the StaphaurexTM Latex Agglutination Test (ThermoFisher, Waltham, Germany).

Detection of Listeria monocytogenes

Enrichment for *L. monocytogenes* was done by mixing 25 g of each food sample with 225 ml of *Listeria* enrichment broth (Biolab, South Africa), homogenized, and incubated at 37 °C for 21 h (ISO, 2017). Thereafter, 200 μ l was plated on prepared RAPID' *Listeria* spp. Agar (Biolab, South Africa) plates containing 0.01 g/L pimaricin (Promepharma, South Africa) and incubated at 37 °C for 24 h. Plates showing blue to blue-green colonies were recorded as positive plates. The identities of colonies were confirmed by means of a latex agglutination test using the OxoidTM Listeria Test Kit (Zymo Research, Irvine, USA).

Identification of bacteria by 16S rRNA gene sequencing

For genomic DNA extraction, individual colonies with different morphology were isolated from incubated plate count agar plates of each ready-to-eat SVF sample, spread on freshly prepared plate count agar plates, and incubated at 37 °C for 21 h. Genomic DNAs were extracted from the harvested cells of individual colony cultures using a ZR Fungal/Bacterial DNA MiniPrepTM Kit (Zymo Research Irvine, USA) and the quantity was ascertained using a NanoPhotometer[®] P-Class P360 version 2.1 (IMPLEN, Schatzbogen, Germany).

Prior to 16S rRNA gene sequencing, the primers ENV1 (5'-AGA GTT TGA TII TGG CTC AG-3') and ENV2 (5'-CGG ITA CCT TGT TAC GAC TT-3'), synthesized by Inqaba Biotechnology Company (Pretoria, South Africa) and correspond to position 8-27 and 1511-1492 of E. coli 16S rRNA, were used to amplify the 16S rRNA. The reaction of each sample was made up with the specific primer pair (1 µM each primer), 2X Polymerase Chain Reaction (PCR) Master Mix (Promega, Madison, USA), 100 ng of DNA template, and nuclease free water up to 25 µl. The amplification was done using a Mx3005P qPCR system (Agilent Technologies, Waldbronn, Germany) with an initial denaturation of 2 min at 94 °C, followed by 30 cycles of denaturation (15 s at 95 °C), annealing (30 s at 48 °C), and elongation (30 s at 72 °C). This was followed by a last final extension at 72 °C for 10 min (Olofsson et al., 2007).

Agarose gel (1.5%) electrophoresis was done for assessing the amplification using the Mini-Sub[®] Cell GT

(Bio-Rad Laboratories Inc., California, USA) and visualized using the GelDoc-ItTM 310 Imaging System (UVP, Cambridge, United Kingdom). Each successful amplified PCR product was purified by MinElute PCR Purification Kit (Whitehead Scientific Ltd, Johannesburg, South Africa) based on the manufacturer's protocol. Inqaba Biotechnology Company (Pretoria, South Africa) conducted the sequence of 16S rRNA amplicons. The sequences were subsequently manually edited using ClustalW in BioEdit (vision 7.0.9.1). The edited sequences were then blasted in the EMBL nucleotide sequence database and species identification was considered at sequence similarity \geq 97% (Stackebrandt and Goebel, 1994).

Data analysis

All bacterial counts were transformed to log10 CFU/g of food sample prior to statistical analysis. The guidelines of Food Safety Authority of Ireland (FSAI, 2016) and South Africa's Foodstuffs, Cosmetics and Disinfectants Act, 54 of 1972 (DOH, 2011) were used to interpret the microbiological quality of ready-to-eat SVFs. The aerobic colony counts were assessed as follows: satisfactory (<3 log CFU/g of sample), borderline (≥ 3 to <5 log CFU/g of sample), and unsatisfactory ($\geq 5 \log CFU/g$ of sample). The Enterobacteriaceae counts were assessed as follows: satisfactory (<2 log CFU/g of sample), borderline ($\geq 2 \leq 4$ log CFU/g), and unsatisfactory (>4 log CFU/g of sample). All pathogen counts were assessed as follows: satisfactory (absence per 25 g of sample) and unsatisfactory (presence per 25 g of sample). Chi-Square was used to analyze the relationship between the microbial quality assessment outcome and SVF groups and significance was considered at $p \le 0.05$. Data analysis was done using IBM SPSS Statistics V25.0.

Results

Totally, 85.37% of the 205 ready-to-eat SVF samples had aerobic growth. Flavored popcorn, fried hake, cheeseburgers, samosas, and hotdog sausages had the highest percentage of aerobic colony counts with above 4 log CFU/g (Table 2). The average aerobic plate mean counts of all ready-to-eat street-vended food categories ranged from 3.4 to 4.12 log CFU/g. There was no significant (p=0.082) association between the aerobic colony count assessment outcomes and SVF groups. Overall, the majority (80%) of ready-to-eat SVF samples had a borderline aerobic colony count while the minority (20%) had unsatisfactory count. None of the samples had unsatisfactory aerobic colony count (Table 3).

Enterobacteriaceae growth was found in 78.18% of the 110 examined SVF samples. All samples within the sandwich-based food category were contaminated

with Enterobacteriaceae (Table 4). There was no significant (p=0.646) association between the Enterobacteriaceae quality assessment outcomes and SVF groups. Overall, 14.5, 70.9, and 14.6% of the SVF samples had unsatisfactory, borderline, and satisfactory Enterobacteriaceae counts, respectively (Table 5).

Among the 272 aerobic growth bacteria isolates, the most frequently genus in the different food samples was *Pseudomonas* (10.66%). The most abundant identified species were *Pseudomonas aeruginosa*, *Escherichia vulneris*, *Stenotrophomonas maltophilia*, *Ralstonia pickettii*, *Bacillus pimulus*, *Enterococcus brevis*, and *Enterococcus faecium*, respectively (Table 6).

Out of the 110 SVF samples, the prevalence rates of *L.* monocytogenes, *S. aureus*, *Salmonella* spp, and *E. coli* O15:H7 were 46.36, 31.8, 21.8, and 1.8%, respectively. *L. monocytogenes* was mainly detected in 60% of starch-based, 52% of beef-based (52%), and 48% of sandwich-based foods, whereas *E. coli* O15:H7 was detected only in 8% of beef-based foods (Table 7). Statistical analyses revealed that there was no significant difference in the prevalence rates of *L. monocytogenes* (p=0.554), *S. aureus* (p=0.137), *Salmonella* spp. (p=0.443), and *E. coli* O15:H7 (p=0.639) in the various SVF groups.

Discussion

In the current survey, the majority of ready-to-eat SVFs had borderline counts (below the 10^5 CFU/g) which are the maximum acceptable limits prescribed by the Food Safety Authority of Ireland (FSAI, 2016) for ready-to-eat foods on the market. The high number of borderline aerobic colony counts is indicator of poor food hygiene practices and sanitary conditions at street food vending sites. The finding of this research is similar to that conducted in Ethiopia (Amare et al., 2019) and Bangladesh (Hossain and Dey, 2019), in which the aerobic colony count of SVFs were found to range of 1×10^4 -1.86×10⁵ CFU/g and 1.2×10^3 - 4.2×10^9 , respectively. Furthermore, in study conducted in Porto region, Portugal, all SVF samples were found to possess aerobic colony counts above 10⁵ CFU/g (Campos et al., 2015). The weak food safety training and the lack of awareness of proper sanitary practices are the main root causes of such high contamination levels. Borderline counts imply that environmental health officials in the Johannesburg Metropolis should investigate the reasons for such counts and take progressive action to reduce them to satisfactory levels. It is worth noting that most informal food business operators in the developing countries are inadequately regulated and the inspections of street food vending sites are often poorly coordinated.

The high level of Enterobacteriaceae counts in readyto-eat SVFs of this survey is an indication of unhygienic production processes which facilitate post-processing contamination. Higher Enterobacteriaceae count were found in a study conducted in Portugal in which all street vended food samples were found to have unsatisfactory Enterobacteriaceae count (Campos et al., 2015). Similarly, Kotzekidou (2013) reported that 35.3% of readyto-eat foods and ready-to-bake frozen pastries from university canteens in Greece were contaminated with Enterobacteriaceae, ranging from 10^3 to 10^4 CFU/g. Another study in Spain showed that less than 30% of plant-based foods sampled from food service establishments contained unacceptable Enterobacteriaceae levels (Sospedra et al., 2013).

The genus detected most frequently in the SVF samples in the current research was Pseudomonas, followed by Escherichia, Bacillus, Stenotrophomonas, Ralstonia, and Lactobacillus. Most of the predominant genera are psychotrophs, which are ubiquitously distributed in water, soil, and the plant and animal environment, explaining their prevalence in SVFs prepared under inadequate hygiene conditions (Rajmohan et al., 2002). A study from Beijing, China indicated that these bacteria were dominant in the microbial community of prepared foods in the supermarkets (Wang et al., 2019). Some species such as P. aeruginosa as well as E. vulneris can become an opportunistic pathogen (Lister et al., 2009). The dominance of species of P. aeruginosa, E. vulneris, and B. pimulus in the studied SVF samples can be attributed to a lack of proper food hygiene practices during handling by street food vendors. These bacterial species, which often do not constitute a risk to public health, can cause opportunistic infection in humans and spoil a wide range of foodstuffs (Caldera et al., 2016; Jain et al., 2016; Yuan and Gao, 2015).

In the present work, L. monocytogenes was the most identified food-borne pathogens in ready-to-eat SVFs, followed by S. aureus, Salmonella spp., and E. coli O15:H7. Listeria has increasingly become a food-borne pathogen of interest, because it is widespread in foods and in the environment. It can also contaminate the food-processing environment as well as ready-to-eat foods (Todd and Notermans, 2011). The fact that it was detected in 20% of ready-to-eat food samples in this survey implies that it constitutes a health risk to consumers. Similarly, a prevalence of 11% was recorded for L. monocytogenes in ready-to-eat meats in Windhoek, Namibia (Shiningeni et al., 2019). In addition, a prevalence of 32.14% was observed for ready-to-eat seafood samples in the Veneto Region, Italy (Gambarin et al., 2012).

Table 1: Description of ready-to-eat street vended foods sampled in the current study

Starch-based foods

Corn flour porridge (pap): a thick maize porridge made with maize flour Fried potato chips: fried finger chips Cookies: baked, sweetened and flattened white flour dough, cut into small pieces Chocolate cookie: baked, sweetened, chocolate-flavoured and flattened flour dough shaped into small pieces Steamed bread: bread dough baked in a bowl inside a pot containing water Bean cake: baked dough made with bean flour Roasted peanuts: unshelled, salted peanuts dried in a frying pan Boiled peanuts: unshelled, salted peanuts boiled in water Flavoured popcorn (skopas): coloured and flavoured popcorn Bean stew: thick, saucy bean soup Peanut flour soup: soup made with peanut flour Boiled maize: fresh corn on the cob, boiled in water Grilled maize: fresh corn on the cob, grilled on a charcoal fire Roasted, shelled peanuts: peanuts, without shells, roasted in a dry frying pan Biscuits: locally made hard, flat and unleavened baked product from wheat flour **Beef-based foods** Bologna sausage (polony): sliced pieces of sausage used to make sandwiches Stewed beef bones: stew, prepared using beef bones Beef stew: stew, prepared using small pieces of beef Beef broth: soup consisting of meat cooked in a stock Boiled beef: spiced, large chunks of beef, boiled in a little water Fried beef tripe: stomach of a cow, fried in vegetable oil Fried ox liver: ox liver fried in vegetable oil Barbecued sausage: sausage grilled on a charcoal fire Beef biltong: cured pieces of dried meat Poultry-based foods Grilled chicken gizzards: chicken gizzards grilled on a charcoal fire Boiled egg: eggs boiled in water Fried chicken pieces: chicken pieces fried in vegetable oil Chicken stew: stew, prepared using small pieces of chicken Grilled chicken feet: whole chicken feet grilled on a charcoal fire Grilled chicken neck: whole chicken necks grilled on a charcoal fire Fish-based foods Fried hake: hake pieces fried in vegetable oil Fried snoek: snoek pieces fried in vegetable oil Vegetable-Based Foods Vegetable soup: broth containing tomato juice, water, potatoes, carrots, celery, undrained chopped tomatoes, green beans and corn Pickled mango (atchaar): spiced and pickled unripe mango pieces Vegetable relish (chakalaka): spicy vegetable dish of onions, tomatoes, chilli and (often) beans Mixed vegetable salad: salad consisting mostly of lettuce, cucumber and tomato Sandwich-based foods Cheese burger: bread roll filled mostly with a cheese patty Cheese/egg burger: bread roll filled mostly with a cheese patty and topped with a poached egg/omelette Samosa: a savoury pastry containing spiced vegetables and/or meat fried in oil Bunny chow: popular in the townships of Gauteng, made by filling a hollowed-out quarter loaf of bread with ingredients such as fried chips,

cheese, polony and pickled raw mango pieces

Hotdog sausage: hotdog bun filled with a grilled sausage

Like the findings of this study, 34.2% of raw salmon finger sushi sold in Hong Kong was contaminated with *S. aureus* (Liang et al., 2016). Also, presence of *Salmonella* spp. was reported in 39% of barbecued pork samples in Hong Kong (Ng et al., 2013), 19.7% of ready-to-eat SVFs in Eastern Ethiopia (Bereda et al., 2016), and 20.8% of raw chicken meat at retail markets in Malaysia (Thung et al., 2016). The presence of these bacterial pathogens in cooked SVFs can be attributed primarily to the post-processing contamination.

Table 2: Total aerobic colon	y counts in ready-to-eat street-ve	ended foods sold in the Johannesburg Metropolis, South Africa
------------------------------	------------------------------------	---

Food groups	Number of positive samples	Average counts (log CFU/g)	
	(%)		
Starch-based foods (n=75)	53 (70.67)	3.40±0.71	
Cornflour porridge (pap) (n=5)	4	3.20±0.43	
Fried potato chips (n=5)	2	4.15±0.35	
Cookies (n=5)	3	2.01±0.23	
Chocolate cookie (n=5)	5	2.97 ± 0.05	
Steamed bread (n=5)	5	3.07±0.67	
Bean cake (n=5)	5	3.88 ±0.40	
Roasted peanuts (n=5)	2	2.57 ± 0.04	
Boiled peanuts (n=5)	5	4.16±0.38	
Flavoured popcorn (skopas) (n=5)	5	4.33±0.45	
Bean stew (n=5)	1	3.79±0.23	
Peanut flour soup (n=5)	3	3.46±0.30	
Boiled maize (n=5)	3	3.73±0.26	
Grilled maize (n=5)	4	3.53±0.30	
Roasted, shelled peanuts (n=5)	2	2.65±0.07	
Biscuits (n=5)	4	3.56±0.36	
Beef-based foods (n=45)	39 (86.67)	3.43±0.55	
Bologna sausage (polony) (n=5)	5	4.00±0.42	
Stewed beef bones (n=5)	5	4.12±0.39	
Beef stew (n=5)	4	3.68±0.24	
Beef broth $(n=5)$	5	3.08±0.24	
Boiled beef (n=5)	5	3.48±0.28	
Fried beef tripe (n=5)	3	2.74±0.23	
Fried ox liver (n=5)	4	3.41±0.07	
Barbecued sausage (n=5)	3	2.63±0.11	
Beef biltong (n=5)	5	3.70±0.36	
Poultry-based foods (n=30)	28 (93.33)	3.56±0.55	
Grilled chicken gizzards (n=5)	5	4.13±1.04	
Boiled egg (n=5)	5	3.27±0.36	
Fried chicken pieces (n=5)	5	3.51±0.17	
Chicken stew $(n=5)$	4	3.65 ± 0.41	
Grilled chicken feet (n=5)	5	3.74±0.65	
Grilled chicken neck (n=5)	4	3.02±0.03	
Fish-based foods (n=10)	10 (100)	4.12±0.57	
Fried hake (n=5)	5	4.58±0.22	
Fried snoek (n=5)	5	3.66±0.28	
Vegetable-based foods (n=20)	20 (100)	3.91±0.54	
Vegetable soup (n=5)	5	3.44 ± 0.42	
Pickled mango (<i>atchaar</i>) (n=5)	5	2.57±0.18	
Vegetable relish (<i>chakalaka</i>) $(n=5)$	5	4.76 ± 0.28	
Mixed vegetable salad (n=5)	5	4.89±0.17	
Sandwich-based foods (n=25)	25 (100)	3.95±0.45	
Cheese burger (n=5)	5	4.12±0.28	
Cheese/egg burger $(n=5)$	5	4.12±0.28 3.77±0.30	
Samosa (n=5)	5	4.32±0.30	
Bunny chow (<i>kota</i>) (n=5)	5	4.32±0.30 3.32±0.06	
Hotdog sausage (n=5)	5	5.52±0.06 4.22±0.47	
	-		
Total (n=205)	175 (85.37)	3.73±0.55	

Table 3: Relationship between aerobic colony count assessment outcomes and ready-to-eat street-vended food groups using Chi-Square test

Food groups	Aerobic colony count quality assessment			
	Satisfactory No. (%)	Borderline No. (%)	Unsatisfactory No. (%)	p value
Starch based foods (n=75)	27 (32)	51 (68)	0 (0)	
Beef-based foods (n=45)	12 (26.7)	33 (73.3)	0 (0)	
Poultry-based foods (n=30)	0 (0)	30 (100)	0 (0)	
Fish-based foods (n=10)	0 (0)	10 (100)	0 (0)	0.082
Vegetable-based foods (n=20)	5 (25)	15 (75)	0 (0)	
Sandwich-based foods (n=25)	0 (0)	25 (100)	0 (0)	
Total (n=205)	41 (20)	164 (80)	0 (0)	

-Satisfactory: <3 log CFU/g; Borderline: ≥3 to <5 log CFU/g; Unsatisfactory: ≥5 log CFU/g

Table 4: Enterobacteriaceae counts in ready-to-eat street-vended foods sold in the Johannesburg Metropolis, South Africa

Food groups	Number of positive samples	Average counts	
	(%)	(log CFU/g)	
Starch-based foods (n=20)	18 (90)	3.71±0.69	
Boiled peanuts (n=5)	5	4.21±0.50	
Bean porridge (n=5)	3	4.36±0.57	
Grilled maize (n=5)	5	3.12±0.20	
Biscuits (n=5)	5	3.14±0.31	
Beef-based foods (n=25)	15 (60)	3.07±0.96	
Bologna sausage (polony) (n=5)	3	2.98 ± 1.07	
Beef broth (n=5)	3	2.31±0.23	
Beef stew (n=5)	3	4.36±0.06	
Grilled beef (n=5)	3	3.12±0.19	
Fried ox liver (n=5)	3	3.78±0.29	
Poultry-based foods (n=20)	15 (75)	2.70±1.21	
Boiled eggs (n=5)	3	2.97±0.05	
Fried chicken (n=5)	4	3.93±0.09	
Chicken stew (n=5)	5	2.08±0.14	
Grilled chicken (n=5)	3	1.39±0.13	
Fish-based foods (n=10)	6 (60)	3.71±0.40	
Fried hake (n=5)	3	3.41±0.03	
Fried snoek (n=5)	3	3.85±0.47	
Vegetable-based foods (n=10)	7 (70)	2.66±1.47	
Vegetable relish (chakalaka) (n=5)	4	3.92±0.09	
Mixed vegetable salad (n=5)	3	1.39±0.13	
Sandwich-based foods (n=25)	25 (100)	3.71±0.74	
Cheese burger (n=5)	5	$2.98{\pm}1.01$	
Cheese/egg burger (n=5)	5	2.78±0.47	
Samosa (n=5)	5	4.03±0.44	
Bread fillings (kota) (n=5)	5	3.13±0.31	
Hotdog sausage (n=5)	5	4.33±0.31	
Total (n=110)	86 (78.18)	3.26±0.74	

Table 5: Relationship between Enterobacteriaceae count assessment outcomes and ready-to-eat street-vended food groups using Chi-Square test

Food groups	Aerobic colony count quality assessment			
	Satisfactory No. (%)	Borderline No. (%)	Unsatisfactory No. (%)	P value
Starch based foods (n=20)	0 (0)	15 (75)	10 (25)	
Beef-based foods (n=25)	3 (12)	19 (76)	3 (12)	
Poultry-based foods (n=20)	8 (40)	10 (50)	2 (10)	
Fish-based foods (n=10)	0 (0)	7 (70)	3 (30)	0.646
Vegetable-based foods (n=10)	5 (50)	5 (50)	0 (0)	
Sandwich-based foods (n=25)	0 (0)	22 (88)	3 (12)	
Total (n=110)	16 (14.6)	78 (70.9)	16 (14.5)	

-Satisfactory: <2 log CFU/g ; Borderline: ≥2-≤4 log CFU/g ; Unsatisfactory: >4 log CFU/g

Table 6: The most predominant bacterial genera and their identified species in street-vended foods in Johannesburg, South Africa

Number of predominant bacteria genus (%)	Number of the most abundant species within genus (%)
Pseudomonas: 29 (10.66)	P. aeruginosa: 21 (72.41)
Escherichia: 25 (9.19)	<i>E. vulneris</i> : 19 (76)
Bacillus: 23 (8.46)	B. pimulus: 17 (73.91)
Stenotrophomonas: 21(7.72)	S. maltophilia: 13 (61.90)
Ralstonia: 19 (6.99)	<i>R. pickettii</i> : 15 (79.95)
Lactobacillus: 15 (5.51)	L. brevis: 9 (60)
Enterococcus: 13 (4.78)	E. faecium: 7 (53.85)
Pantoea: 10 (3.68)	P. ananatis: 4 (40)
Weissella:7 (2.57)	W. cibaria: 3 (42.86)
Citrobacter: 7 (2.57)	C. freundii: 4 (57.14)
Leuconostoc: 5 (1.84)	L. mesenteroides: 2 (40)
Serratia: 4 (1.47)	S. proteamaculans: 2 (50)
Others: 94 (34.56)	-
Total: 272 (100)	

Food groups	<i>E. coli</i> O15:H7 No. (%)	Salmonella spp. No. (%)	S. aureus No. (%)	L. monocytogenes No. (%)
Starch-based foods (n=20)	0 (00)	3 (15)	8 (40)	12 (60)
Boiled peanuts (n=5)	ND	1	2	5
Beans porridge (n=5)	ND	1	3	3
Grilled maize (n=5)	ND	1	3	4
Biscuits (n=5)	ND	ND	ND	ND
Beef-based foods (n=25)	2 (8)	5 (20)	12 (48)	13 (52)
Bologna sausage (polony) (n=5)	1	2	4	5
Beef broth $(n=5)$	1	2	3	4
Beef stew (n=5)	ND	1	4	2
Grilled beef (n=5)	ND	ND	1	1
Fried ox liver (n=5)	ND	ND	ND	ND
Poultry-based foods (n=20)	0 (00)	7 (35)	5 (25)	8 (40)
Boiled eggs (n=5)	ND	3	3	2
Fried chicken (n=5)	ND	2	1	3
Chicken stew (n=5)	ND	1	1	2
Grilled chicken (n=5)	ND	1	ND	1
Fish-based foods (n=10)	0 (00)	1 (10)	1 (10)	3 (30)
Fried hake (n=5)	ND	1	ND	2
Fried snoek (n=5)	ND	ND	1	1
Vegetable-based foods (n=10)	0 (00)	1 (10)	1 (10)	3 (30)
Vegetable relish (chakalaka) (n=5)	ND	ND	ND	2
Mixed vegetable salad (n=5)	ND	1	1	1
Sandwich-based foods (n=25)	0 (00)	7 (28)	8 (32)	12 (48)
Cheese burger (n=5)	ND	2	3	5
Cheese/egg burger (n=5)	ND	2	2	3
Samosa (n=5)	ND	1	2	2
Bread fillings (kota) (n=5)	ND	ND	ND	1
Hotdog sausage (n=5)	ND	2	1	1
Total (n=110)	2 (1.8)	24 (21.8)	35 (31.8)	51 (46.36)

Table 7: Prevalence of *Escherichia coli*, *Salmonella* spp., *Staphylococcus aureus*, and *Listeria monocytogenes* in ready-to-eat street-vended foods sold in the Johannesburg Metropolis, South Africa

ND: Not Detected

Conclusion

Based on the findings of this study, the microbial quality and safety of ready-to-eat SVFs sold in the Johannesburg Metropolis remain a serious public health concern. Hence, it is necessary to educate street food vendors and enforce food safety legislation in the street food sector in the country.

Author contributions

C.V.A., S.L.L., as well as F.T.T. designed the research outline, analyzed the data and wrote the manuscript; C.V.A. conducted the experiment. All authors read and approved the final manuscript.

Conflicts of interest

All authors declare that there was no conflict of interest regarding the publication of this article.

Acknowledgements

We acknowledge the Department of Research and Innovation of the University of South Africa for providing the funds required for this study. The research was approved (Code Number: 2014CAES036) by the ethics committee of the College of Agriculture and Environmental Science, University of South Africa.

References

- Alimi B.A. (2016). Risk factors in street food practices in developing countries: a review. Food Science and Human Wellness. 5: 141-148. [DOI: 10.1016/j.fshw.2016.05.001]
- Amare A., Worku T., Ashagirie B., Adugna M., Getaneh A., Dagnew M. (2019). Bacteriological profile, antimicrobial susceptibility patterns of the isolates among street vended foods and hygienic practice of vendors in Gondar town, Northwest Ethiopia: a cross sectional study. *BMC Microbiology*. 19: 120. [DOI: 10.1186/s12866-019-1509-4]
- Asiegbu C.V., Lebelo S.L., Tabit F.T. (2016). The food safety knowledge and microbial hazards awareness of consumers of ready-to-eat street-vended food. *Food Control.* 60: 422-429. [DOI: 10.1016/j.foodcont.2015.08.021]

- Bereda T.W., Emerie Y.M., Reta M.A., Asfaw H.S. (2016). Microbiological safety of street vended foods in Jigjiga City, Eastern Ethiopia. *Ethiopian Journal of Health Science*. 26: 163-172. [DOI: 10.4314/ejhs.v26i2.10]
- Caldera L., Franzetti L., Van Coillie E., De Vos P., Stragier P., De Block J., Heyndrickx M. (2016). Identification, enzymatic spoilage characterization and proteolytic activity quantification of *Pseudomonas* spp. isolated from different foods. *Food Microbiology*. 54: 142-153. [DOI: 10.1016/j.fm.2015.10.004]
- Campos J., Gil J., Mourão J., Peixe L., Antunes P. (2015). Readyto-eat street-vended food as a potential vehicle of bacterial pathogens and antimicrobial resistance: an exploratory study in Porto region, Portugal. *International Journal of Food Microbiology*. 206: 1-6. [DOI: 10.1016/j.ijfoodmicro.2015.04. 016]
- Department of Health (DOH). (2011). Regulations governing microbiological standards for foodstuffs and related matters as amended by Government Gazette 34582 on 2 September 2011. Department of Health, Republic of South Africa.
- Food Safety Authority of Ireland (FSAI). (2016). Guidelines for the interpretation of results of microbiological testing of ready-toeat foods placed on the market. 2nd revision. Guidance Note No. 3.
- Gambarin P., Magnabosco C., Losio M.N., Pavoni E., Gattuso A., Arcangeli G., Favretti M. (2012). *Listeria monocytogenes* in ready-to-eat seafood and potential hazards for the consumers. *International Journal of Microbiology*. [DOI: 10.1155/2012/ 497635].
- Hossain M., Dey B.K. (2019). Microbial contamination of handmade sauce used by street food vendors in Jashore, Bangladesh. *Journal Food Quality and Hazards Control.* 6 :115-120. [DOI: 10.18502/jfqhc.6.3.1385]
- International Organization for Standardization (ISO). (2001). Microbiology of food and animal feeding stuffs-Horizontal method for the detection of *Escherichia coli* 0157. Standard No. 16654.
- International Organization for Standardization (ISO). (2002). Microbiology of food and animal feeding stuffs-Horizontal method for the detection of *Salmonella* spp. Standard No. 6579.
- International Organization for Standardization (ISO). (2003). Microbiology of food and animal feeding stuffs-Horizontal method for the enumeration of coagulase-positive staphylococci (*Staphylococcus aureus* and other species)-Part 3: detection and MPN technique for low numbers. Standard No. 6888-3.
- International Organization for Standardization (ISO). (2004). Microbiology of food and animal feeding stuffs-Horizontal methods for the detection and enumeration of Enterobacteriaceae-Part 2: colony-count method Standard No. 21528-2.
- International Organization for Standardization (ISO). (2013). Microbiology of the food chain -horizontal method for the enumeration of microorganisms-Part 2: colony count at 30 degrees C by the surface plating technique. Standard No. 4833-2.
- International Organization for Standardization (ISO). (2017). Microbiology of the food chain-Horizontal methods for the detection and enumeration of *Listeria monocytogenes* and *Listeria* spp. Part 1: detection method. Standard No. 11290-1.
- Jain S., Nagarjuna D., Gaind R., Chopra S., Debata P.K., Dawar R., Sardana R., Yadav M. (2016). *Escherichia vulneris*: an unusual cause of complicated diarrhea and sepsis in an infant. A case report and review of literature. *New Microbes and New Infections*. 13: 83-86. [DOI: 10.1016/j.nmni.2016.07.002]
- Khongtong J., Ab Karim S., Othman M., Bolong J. (2014). Consumption pattern and consumers' opinion toward street food in Nakhon Si Thammarat Province, Thailand. *International Food Research Journal*. 21: 125-130.
- Kibret M., Tadesse M. (2013). The bacteriological safety and antimicrobial susceptibility of bacteria isolated from street-vended white lupin (*Lupinus albus*) in Bahir Dar, Ethiopia. *Ethiopian Journal of Health Sciences*. 23: 19-26.
- Kothe C.I., Schild C.H., Tondo E.C., Malheiros P.S. (2016). Microbiological contamination and evaluation of sanitary conditions

of hot dog street vendors in Southern Brazil. *Food Control.* 62: 346-350. [DOI: 10.1016/j.foodcont.2015.11.005]

- Kotzekidou P. (2013). Microbiological examination of ready-to-eat foods and ready-to-bake frozen pastries from university canteens. *Food Microbiology*. 34: 337-343. [DOI: 10.1016/j. fm.2013.01.005]
- Liang W.L., Pan Y.L., Cheng H.L., Li T.C., Yu P.H.F., Chan S.W. (2016). The microbiological quality of take-away raw salmon finger sushi sold in Hong Kong. *Food Control.* 69: 45-50. [DOI: 10.1016/j.foodcont.2016.04.015]
- Lister P.D., Wolter D.J., Hanson N.D. (2009). Antibacterialresistant *Pseudomonas aeruginosa*: clinical impact and complex regulation of chromosomally encoded resistance mechanisms. *Clinical Microbiology Reviews*. 22: 582-610. [DOI: 10.1128/CMR.00040-09]
- National Institute of Communicable Diseases (NICD). (2018). Situation update on listeriosis outbreak, South Africa. National Institute of Communicable Diseases, South Africa.
- Ng Y.F., Wong S.L., Cheng H.L., Yu P.H.F., Chan S.W. (2013). The microbiological quality of ready-to-eat food in Siu Mei and Lo Mei shops in Hong Kong. *Food Control.* 34: 547-553. [DOI: 10.1016/j.foodcont.2013.05.018].
- Oguttu J.W., McCrindle C.M.E., Makita K., Grace D. (2014). Investigation of the food value chain of ready-to-eat chicken and the associated risk for staphylococcal food poisoning in Tshwane metropole, South Africa. *Food Control.* 45: 87-94. [DOI: 10.1016/j.foodcont.2014.04.026]
- Olofsson T.C., Ahrné S., Molin G. (2007). The bacterial flora of vacuum-packed cold-smoked salmon stored at 7 °C, identified by direct 16S rRNA gene analysis and pure culture technique. *Journal of Applied Microbiology*. 103: 109-119. [DOI: 10.1111/j.1365-2672.2006.03216.x]
- Rajmohan S., Dodd C.E.R., Waites W.M. (2002). Enzymes from isolates of *Pseudomonas fluorescens* involved in food spoilage. *Journal of Applied Microbiology*. 93: 205-213. [DOI: 10.1046/j.1365-2672.2002.01674.x]
- Rane S. (2011). Street vended food in developing world: hazard analyses. *Indian Journal of Microbiology*. 51: 100-106. [DOI: 10.1007/s12088-011-0154-x]
- Shiningeni D., Chimwamurombe P., Shilangale R., Misihairabgwi J. (2019). Prevalence of pathogenic bacteria in street vended ready-to-eat meats in Windhoek, Namibia. *Meat Science*. 148: 223-228. [DOI: 10.1016/j.meatsci.2018.05.014]
- Sospedra I., Rubert J., Soriano J.M., Mañes J. (2013). Survey of microbial quality of plant-based foods served in restaurants. *Food Control.* 30: 418-422. [DOI: 10.1016/j.foodcont.2012. 08.004]
- Stackebrandt E., Goebel B.M. (1994). Taxonomic note: a place for DNA-DNA reassociation and 16S rRNA sequence analysis in the present species definition in bacteriology. *International Journal of Systematic Bacteriology*. 44: 846-849. [DOI: 10.1099/00207713-44-4-846]
- Thung T.Y., Mahyudin N.A., Basri D.F., Wan Mohamed Radzi C.W., Nakaguchi Y., Nishibuchi M., Radu S. (2016). Prevalence and antibiotic resistance of *Salmonella* Enteritidis and *Salmonella* Typhimurium in raw chicken meat at retail markets in Malaysia. *Poultry Science*. 95:1888-1893. [DOI: 10.3382/ps/pew144]
- Todd E.C.D., Notermans S. (2011). Surveillance of listeriosis and its causative pathogen, *Listeria monocytogenes. Food Control.* 22: 1484-1490. [DOI: 10.1016/j.foodcont.2010.07.021]
- Wang P., Hu A., Fan X., Zhao X., Ge Y., Chen Y. (2019). Bacterial communities in prepared foods available at supermarkets in Beijing, China. *Food Research International*. 120: 668-678. [DOI: 10.1016/j.foodres.2018.11.024]
- World Health Organization (WHO). (2018). Listeriosis-South Africa. Disease outbreak news. World Health Organization, Geneva.
- Yuan Y., Gao M. (2015). Genomic analysis of a ginger pathogen Bacillus pumilus providing the understanding to the pathogenesis and the novel control strategy. Scientific Reports. 5: 10259. [DOI: 10.1038/srep10259]