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FOOD SAFETY PRACTICES AMONG FOOD CARTS IN -NORTH LEBANON

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FOOD SAFETY PRACTICES AMONG FOOD CARTS IN -NORTH LEBANON

Abstract

Street food carts serve different types of popular and traditional foods; it is a common economic sector worldwide. In North Lebanon, food carts are mobile or centered in specific places serving common and well known RTE meals. In addition, the increasing incidence of foodborne illnesses associated with street foods sheds light on the importance of inspecting the practices of street vendors. Therefore, the aim of this study is to assess the food safety in 30 food carts using an observational checklist and to perform microbiological analysis for the detection of foodborne pathogens namely; yeast, mold, *Listeria monocytogenes*, *Salmonella*, Enterohemorrhagic *E.coli*, B-glucuronidase *E.coli*, Enterobacteriaceae and *Clostridium perfringens* in 10 samples of orange juice, 10 samples of cheese caek and 10 samples of meat shawarma; moreover to investigate the prevalence of Methicillin-resistant *Staphylococcus aureus* in RTE foods and street vendor's hands. Out of 30 vendors, 100% were males, 80% with an age between 25-44, 80% of them had less than high school level and 66.7% had fixed stalls. When vendors were observed for food safety practices, 70% and 86.7% of them did not store raw materials separately nor in suitable form respectively, 43.5% cleaned their carts monthly, 96% shared utensils between many types of food, 70% did not clean the counter top surface before starting food preparation; moreover 96%, 76 % and 83% of vendors were not wearing net, gloves nor an appropriate uniform, respectively. Results showed that all tested samples were contaminated with at least one of the tested foodborne pathogens, unsatisfactory levels of yeast and mold were recorded in 10 and 9 orange juice samples, respectively. On the other hand, B-glucuronides *E.coli* exceeded standard limit in two samples of RTE food, one shawarma sample did not meet standard with respect to *C. perfringens*, Remarkably, unsatisfactory levels of *S. aureus* were detected in 55 % of RTE samples and in 30 % of vendor's hands. *S. aureus* strains were susceptible to at least one of the used antibiotics, accordingly one isolated from vendor's hand is considered as MRSA. Therefore, the unsatisfactory levels of yeast, mold and *S. aureus* in tested samples were induced by vendor's violation of food safety practices. Consequently, the current proposes to improve the legislation needs to provide safe food for the end consumer.

Keywords

Food Safety, Microbiology, MRSA, Food Street, Swabs, Vendors, Ancient food

1. INTRODUCTION

Street food trade is an ancient practice common in many countries (Cortese, Veiros, Feldman, & Cavalli, 2016) and considered RTE food, immediately prepared by vendors in the roads and served to consumers (Cortese et al., 2016). Street foods became one of the main interests of public health, since hawkers most of the times are uneducated in addition to their lack in the basal knowledge of personal hygiene and food safety practices. A remarkable relationship has been established between street food trade and food safety (Alimi, 2016). According to World Health Organization (WHO) some violation of food street practices were noted, raw materials are of low grade and safety especially most of the time they are not from reliable sources, and the wrong storage practices causes deterioration of food (World Health Organization, 1996). In most countries, street trade is outside government control and follow up, which is one of the causes of their violation in food safety (Alimi, 2016; Cortese et al., 2016). Foodborne diseases are on the rise in many regions due to the uncontrolled increase in the number of untrained hawkers around the world (Manguiat & Fang, 2013). There are many aspects of foodborne illness according to microbial (bacteria, yeast and mold) prevalence related to preparation area, method of cooking, raw components, inadequate food storage conditions. Recent studies correlate the occurrence of foodborne outbreaks to consumption of fruit juices and RTE food (Aneja, Dhiman, Aggarwal, Kumar, & Kaur, 2014). The aim of this study is to assess contamination in street food and improving legislation needed to provide safe food for the end consumer. This was achieved by evaluation of carts using observational checklist, for their knowledge and behaviors in food safety checklist and laboratory analysis of the collected samples. A correlation was achieved between exceeding *S. aureus* and wrong hygienic practices. Moreover, swabs were collected from their hands to assess the prevalence of MRSA in food samples and vendor's hands.

2. METHODOLOGY

2.1 Study Population

The study was conducted in Tripoli, the capital city of North Lebanon. The participants were street food vendors serving fresh orange juice and RTE food namely, cheese cake and shawarma. In total, 30 street food vendors were randomly chosen to fill out a food safety checklist to assess their knowledge, behaviors in food safety practices. Moreover, swabs were collected from their hands to assess the prevalence of MRSA

2.2 Ethical Approval

The participants were informed about the aim of the study and the objectives to be achieved. Moreover, they were informed that their participation would be anonymous and voluntary where their responses were held in a confidential manner.

2.3 Study Tools

Food safety observational checklist.

A checklist was used for the evaluation of the food safety habits of street food hawkers and the vending areas. The checklists (Table 1 & 2) was adopted from previous studies and adjusted to suit the aim of this work (Fssai,2009;Cortese, Veiros, Feldman, & Cavalli, 2016). The checklist has two sections.

Section 1: Demographic data about participants, consisting of 4 questions namely; gender, age, level of education and the vending site type.

Section 2: It consists of 6 major sectors:

(A) information about the point of sale, (B) personal hygiene, (C) Attribute on the final purchased product, (D) left over storage, (E) source of raw material and (F) cleaning of carts

The data was recorded as yes and no, the aim of the research was explained to the hawkers primary and after they responded to participate, an explanation for the different stages of the study were given to every participant before the observation of food preparation and hand swabs collection.

2.4 Data Collection

Between September and November 2020, 30 street food vendors participated to fill out a food safety observational checklist. The vendors were approached randomly in different streets in Tripoli. When the vendors provided us with an oral consent to participate in this study, they were requested to answer the questionnaire and to take swabs from their hands. To save time and avoid bias, the observational checklist was completed by the researcher.

2.5 Sample Collection

A total of 10 fresh orange juice and 20 RTE food samples (10 Orange juice [Sample 1 (S1)-S10], 10 cheese caek [S11-S20] and 10 meat shawarma [S21-S30]) were collected from 30 different vendors in an alternative location in street and sidewalks in Tripoli. Food samples were bought as the same conditions they bought by consumers and collected in sterile plastic bags. After collection, samples were immediately kept in an ice box 4-degree Celsius (°C) and transported to LARI (Lebanese Agricultural Research Institute) laboratory within 3- 4 hours (hrs). to be tested for their microbial load.

2.6 Swab Collection

A total of 20 swabs (10 from cheese caek vendor's hand and 10 from meat shawarma vendor's hand) same vendors of the prospected food samples. The collected swab samples were brought aseptically to their sterile covers and kept in an ice box (4°C) immediately after collection and transported to LARI laboratory within 3-4 hrs. to be tested for the prevalence existence of *S. aureus* and MRSA (Sarfrac A., 2015).

2.7 Statistical Analysis

All Data were analyzed using Statistical Package for Social Science (SPSS) 23.0 and Excel Microsoft 2015. Values were expressed by using mean of bacterial load and expressed in logarithm colony forming units/gram or / milliliter (\log_{10} CFU/g or per (ml)) (Beshiru et al., 2019). Descriptive analysis was performed to calculate the mean, then compared using one-way ANOVA and the significance of differences was considered at 95% confidence interval probability ($p < 0.05$). Mann-Whitney U test was performed to specify a relation between wrong food safety practices of vendors and the RTE food samples that have an exceeding limits of *S. aureus* as recommended by International Standard Organization (ISO) 688-1 :1999 of Lebanese Standards Institution (LIBNOR) standard. A correlation with $p < 0.05$ was considered as significant.

2.8 Microbiological Analysis

2.8.1 Food sample processing and bacterial load

According to Zhang, 2019 a food mixture was prepared by adding 25 g or 25 ml (for juice) of collected samples to 225 ml of Buffered Peptone Water (BPW), then the mixture was homogenized in a stomacher for 30 sec. The prepared solutions of the 30 collected food samples were used as a stock solution for the detection and enumeration of Enterobacteriaceae, B-Glucuronidase *E-coli*, Enterohemorrhagic *E. coli*, *C. perfringens*, *L. monocytogenes*, *Salmonella* spp, Yeast and Molds. The prevalence of *S. aureus* was only tested in food samples of cheese caek and meat shawarma and on street food vendor's hands. For total bacterial count, one ml of the stock solution was spread on Plate Count Agar (PCA) and incubated at 30°C for 72 hr. The number of colonies was counted as CFU per gram of tested samples and expressed as log cfu/g or per ml (Islam et al., 2019), then the average was calculated.

2.8.2 Isolation and identification of foodborne pathogens in food samples

Following the ISO 21528-2017-06 for detection and enumeration of Enterobacteriaceae, 1 ml of each processed sample was pipetted to an unfilled petri dish and then poured with Violet Red Bile Glucose Agar (VRBGA). The plates were incubated aerobically at 37°C for 24 hr. Five colonies were selected and then inoculated on Nutrient Agar (NA) using streaking dilution method, after that the plates were incubated at 37°C to 24 hrs. to obtain pure cultures. For further confirmation, the oxidase test was performed, where a pure selected colony was stricken on the oxidase filter strip and the result was read within 10 sec. Furthermore, glucose fermentation test was performed by stabbing a pure colony into glucose fermentation agar tube then incubated at 37°C for 24 hr. Concerning the detection and enumeration of B-

Glucuronidase *E. coli* in the tested food samples, it was performed according to ISO 16140 using Tryptone-Bile-Glucuronide (TBX) medium, then the plates were incubated at 44°C for 24 hrs. On the other hand, for the identification and enumeration of *C. perfringens*, the ISO 7937-2004 was performed by inoculating 1 ml of sample suspensions to be tested into Tryptase Sulfite Cycloserine (TSC) medium using pour plate technique, then the inoculated plates were incubated anaerobically at 37°C for 24 hrs. For further confirmation of the isolated colonies, Lactose Sulfite (LS) medium was used. Two pure presumptive colonies were inoculated into 10 ml Thioglycolate Medium (TM), then incubated at 37°C for 24 hr. Subsequently, 5 drops of the thioglycolate culture were transferred to the LS medium, then incubated aerobically at 46°C for 24 hr. Each LS broth that showed gas production with black precipitate was further confirmed for the presences of *C. perfringens* using API test kit according to the manufacturer's instructions. On the other hand, only meat shawarma and cheese cake samples were tested for the presence of *S. aureus* according to ISO 688-1 :1999 for the enumeration of coagulase positive staphylococci, where 1 ml of the processed samples was pipetted equally into 3 plates of Baird Parker agar (BP), the plates were inoculated using spreading technique, and incubated for 48 hrs at 37°C. For further confirmation, 5 presumptive colonies were selected then added to diagnostic reagents in a diagnostic paper (rapid latex test). Pure culture was obtained by inoculating 1 colony into Brain-Heart Infusion broth (BHI), then incubated at 37°C for 24 hrs. Furthermore, coagulase test was performed by transferring 0.5 ml of the culture to 0.5 ml rabbit plasma in sterile hemolysis tubes and incubated at 37°C for 24 hrs.

2.8.3 Isolation and Identification of Enterohemorrhagic *E. coli*

According to ISO 16649-2:2001 the sample suspension was prepared by adding 25 ml of orange juice or 25g of cheese caek or meat shawarma to 225 ml Tryptic Soy Broth (TSB) with novobiocin mixture then homogenized in a stomacher bag for 30 sec and incubated at 41.5⁰ C for 24 hr., a loopful of the overnight culture was streaked by dilution method on Chromogenic Agar (CHROM) and incubated at 37°C for 24 hr., 5 violet presumptive colonies are selected for further confirmation using diagnostic reagents in a diagnostic paper (rapid latex test).

2.8.4 Isolation and Identification of *L. monocytogenes*

The isolation and identification of *L. monocytogenes* was performed according to ISO 11290-1:2017-05, the sample suspension was prepared by adding 25 ml of orange juice or 25 g of cheese caek or meat shawarma to 225ml of Half Frazer Broth (HFB), then homogenized in a stomacher bag for 30 sec and incubated at 30°C for 24 hr., then 0.1 ml of culture was added to 10 ml of Listeria Selective Fraser Broth (FB) and incubated at 37°C for 24 hr. One loopful from the incubated culture was inoculated to both Chromogenic Listeria agar and PALCAM agar and streaked by dilution method then incubated for 48 hr. at 37°C. Two green presumptive colonies are selected and inoculated on Tryptone Soya Yeast Extract Agar (TSYEA) at 37°C for 24 hr. For confirmation Hemolysis test is used by single stabbed inoculum from the culture on sheep blood agar then incubated at 37°C for 24 hr. Further confirmation is carried out using API listeria test kit according to the manufacturer's instructions.

2.8.5 Isolation and Identification of *Salmonella* spp

Following ISO 6579 2017-02, 0.1 ml of the homogenized sample suspension prepared by adding 25 ml or 25g of food samples to 225ml of BPW, was added to 10 ml Rappaport Vassiliadis Soy broth (RVS) then incubated at 41.5°C for 24 hr. After that 1ml of the culture was added to 10 ml of Muller Kauffmann Tetra Thionate novobiocin (MKTTn) broth and incubated at 37°C for 24 hr. Then a loopful was immersed in each broth to inoculate both Xylose Lysine Deoxycholate agar (XLD) and Salmonella Shigella agar (SS) agar plates using quadrant method and incubated for 24 hr. at 37°C. For pure culture 2 colonies having black color are selected then streaked using quadrant method on NA, and incubated at 32°C for 24 hr. For further confirmation API 20E is used.

2.8.6 Isolation and Identification of Yeast and Molds

The isolation and identification of yeast and molds was carried out according to ISO 21527-1:2008. A stock sample suspension was prepared by adding 25 ml or 25g of food samples to 225ml of BPW and homogenized for 30 sec in a stomacher. A serial dilution was performed by transferring 1 ml of the initial suspension in 9 ml saline water, then 1 ml of the diluted sample was transferred equally to 3 plates of Dicloran-Rose Bengal Chloramphenicol (DRBC). The inoculated plates were incubated at 25°C for 3 to 5 days.

2.8.7 Isolation and Identification of *Staphylococcus aureus* on Hand Swabs

10 ml of BPW was added to the swabs, then continuous streaks were made on Mannitol Salt Agar (MSA) using swabbing technique, then incubated for 24 hr. at 37°C. For confirmation, coagulase test was performed, 5 colonies were selected and added to diagnostic reagents in a diagnostic paper rapid latex staphylect, for pure culture 1 colony was transmitted to BHI broth then incubated at 37°C for 24 hr. After that 0.5 ml of positive coagulase test results were added aseptically to 0.5 ml rabbit plasma in sterile hemolysis tubes and incubated at 37°C for 24 hr. (El Darra et al., 2019).

2.8.8 Antimicrobial Susceptibility Testing

Susceptibility of the *S. aureus* isolates to some antimicrobial agents was determined using disc diffusion method according to the Clinical Laboratory Standards Institute (CLSI) guidelines (Islam et al., 2019). One pure colony of the isolated *S. aureus* previously streaked on Tryptone Soy Agar (TSA) and incubated at 37°C for 48 hr., was transferred to saline water to achieve 0.5 McFarland turbidity (10^8 cfu/ml). A swab was immersed in prepared saline water then swiped to completely wipe on MHA. The used antimicrobial agents involved (Amoxicillin 25 µg, Cefoxitin 30 µg, Oxacillin 1 µg and Tetracycline 30 µg). The antibiotic discs were aseptically impregnated into agar plates using a sterile forceps and equidistant apart. The inoculated MHA plates were kept at room temperature for 15 min and then incubated at 37°C for 18–24 hr. The susceptibility, intermediate and resistance to antibiotics of *S. aureus* isolates was determined by the measurement of inhibition zone diameters and then compared with the interpretative chart of CLSI. *S. aureus* isolates resistant to Cefoxitin 30 µg were reported as MRSA, however the resistance to the other 3 antibiotics (Amoxicillin 25 µg, Oxacillin 1 µg and Tetracycline 30 µg) reported as Multidrug Resistance (MDR) (CLSI, 2020).

3. RESULTS & DISCUSSION

3.1 Analysis of Street Food Vendor's Profile

In the present study, a total of 30 vending sites were targeted in most crowded areas in North Lebanon, between September and November 2020. Therefore, 30 street vendors participated in this study. All vendors were male (Table 1) which coincides with similar studies in Lebanon (Loukieh et al., 2018), even in Brazil it showed men dominance in 58% (Cortese, Veiros, Feldman, & Cavalli, 2016). However, worldwide researches showed that women predominate this sector (Cortese et al., 2016), similarly in Nigeria (Aluko, Ojeremi, Olaleke, & Ajidagba, 2014), Uganda (Asiegbu et al., 2016; Muyanja et al., 2011) and Ireland (Moreb, Priyadarshini, & Jaiswal, 2017). The results of socio-demographic characteristics of the present study showed that 80% were holding only primary school level (Table 1) less than India's results, where 45% were holding the elementary school degree (Bhattacharjya & Reang, 2014). While a recent study in Lebanon showed that 33.3% of the vendors were holding secondary certificate.

3.2 Assessment of Food Safety Knowledge and Practices Among Food Carts

Lack of water connection at the point of sale was detected in almost 14 carts (Table 2). According to WHO, the absence of connectivity to a water supply in the food carts serving street food is a major source of population health threat, since water is not only used in food as an ingredient, it is used for washing equipment and hands, cleaning and other operations. Moreover, it is well known that in Tripoli, water is not available all the time. Therefore, the

unavailability of water at the point of sale in food carts affects other food safety practices and poses the population health at risk.

The results of this study showed that, in 70% and 86.67% of food carts, raw materials are not stored either separately nor in suitable form respectively (Table 2), which increases the probability to get food deterioration due to temperature changes; moreover, most raw materials of street food are with the lowest grade due to their cheapest price. About 33.3% of the carts are mobile (Table 2), thus they are at microbiological, chemical and physical contamination risk in addition to temperature changes, such conditions made food in the carts unsafe to be consumed, since the majority of street foods were raw and directly prepared, and may remain in the danger zone for long time (Table 2). On the other hand, most food leftover remains in the carts and not refrigerated (Table 2), such conditions increase the risk of bacterial proliferation in the danger zone and leading to serious and life threatening foodborne illness (FSIS, 2011). Mobile carts increase the probability of hazardous contamination of food, even the non-mobile one, due to the long period time the carts got cleaned. In this study 30%, 26.67% and 43.33% of carts were cleaned daily, weekly or monthly (Table 2). The personal hygiene is one of the main factors affecting the safety of food, especially that bacteriological infection may be transmitted from the vendors to the served food (Chukwudiegwu & Ogonna, 2020). In the present study, 96%, 76 % and 83% of vendors were not wearing net, gloves nor an appropriate uniform respectively (Table 2); thus, increasing the risk of transmission of dirt, hair traces and other contaminant to the served food. Moreover, they did not wash their hand between money transaction (Table 2), all these finding were in coincidence with other studies showing that 80% of street vendors did not have any training on food safety practices and were not aware of food safety laws and regulations since money transaction was considered as a vulnerable bacteriological vehicle that can transmit bacteria between vendors and consumers (Gedik, Voss, & Voss, 2013). On the other hand, recent studies showed that street food usually are saturated with fat, energy, salts and sugars, leading to obesity and cardiovascular diseases (Greater et al., 2018); furthermore, 53% of the Lebanese adolescents were considered as obese (Jurjus & Braysh, 2016). According to WHO the obesity in Lebanon is the risk factor that increases the mortality rate higher than tobacco or even blood pressure diseases (WHO-NCD, 2018). Moreover, 47% of the annual deaths in Lebanon are Non-Communicable Disease (NCD) patient (Greater, Area, & Cohort, 2018).

3.3 Microbiological Quality of Served Food by Street Vendors

The business of food carts increasing rapidly in Lebanon. It offers the consumers' needs regarding rapid snack, fresh juice with affordable prices, the specificity of such carts is their location, the fixed carts are located near to the main and shopping centers; moreover, the mobile of them, are movable in the streets and increasing the risk of food contamination due to wrong preparation practices (Hilario, 2015; U. S. Food and Drug Administration, 2013).

In the present study, the microbiological quality of studied food categories was compared to the Lebanese Standards Institution (LIBNOR) for fresh orange juice and RTE food. The results showed the load of yeast and mold in 100% and 90% of the tested samples respectively (Table 3), did not meet standards and exceeded the norms to reach an average of $4.78 \log_{10}$ cfu/ml and $4.18 \log_{10}$ cfu/ml respectively (Figure 1), these findings were higher than other studies showing that the percentage of contaminated samples with unsatisfactory load in India 68% and 52.7% of the tested samples were classified as unsatisfactory for yeast and mold count respectively (Aneja, Dhiman, Aggarwal, Kumar, & Kaur, 2014). On the other hand, Studies showed that yeast, the main cause of fruits spoilage, can grow under low temperature till 4°C (Tournas et al., 2006), these fruits spoilers, such as *Aspergillus flavus*, may increase in number during preservation with mycotoxin production, rendering the consumers with weakened immune system at risk (Aneja et al., 2014)

Table 1: Socio-demographic characteristics of the street food vendors and the type of vending site in Tripoli.

Variables	Number	(%)
Gender		
Male	30	(100 %)
Female	0	(0 %)
Age		
15-24	0	(0%)
25-44	24	(80 %)
> 45	6	(20 %)
Education level		
Elementary school degree	24	(80 %)
High school degree	6	(20 %)
University degree	0	(0 %)

Table 2: Food safety observational checklist of street food vended in Tripoli

Observed parameters	Orange Juice		Cheese caek		Shawarma	
	Yes	No	Yes	No	Yes	No
A. Point of sale						
Clean surface	80% (8)	20% (2)	30% (3)	70% (7)	40% (4)	60% (6)
Connectivity to water supply	60% (6)	40% (4)	0% (0)	100% (10)	100 % (10)	0% (0)
Cross-contamination	60% (6)	40% (4)	60% (6)	40% (4)	100% (10)	0% (0)
Clean, maintained and protected equipment & utensils	40% (4)	60% (6)	100% (10)	90% (9)	100% (10)	0% (0)
Separate equipment & utensil for each food	10% (1)	90% (9)	0% (0)	100% (10)	0% (0)	100% (10)
Equipment's are free of physical defects	40% (4)	60% (6)	70% (7)	30% (3)	100% (10)	0% (0)
Fresh appearance of raw materials	80% (8)	20% (2)	30% (3)	70% (7)	NA	
Suitable storage of raw materials	0% (0)	100% (10)	0% (0)	100% (10)	40% (4)	60% (6)
Storage of raw materials separately	50% (5)	50% (5)	40% (4)	60% (6)	0% (0)	100% (10)
Food packaging meeting standards	100% (10)	0% (0)	0% (0)	100% (10)	100% (10)	0% (0)
Immediate disposal of leftover	60% (6)	40% (4)	90% (9)	10% (1)	NA	
Working surface area are cleaned properly before starting work	60% (6)	40% (4)	30% (3)	70% (7)	0% (0)	100% (10)
Direct food preparation	20% (2)	80% (8)	80% (8)	20% (2)	100% (10)	0% (0)
Vendors appear in good health	70% (7)	30% (3)	60% (6)	40% (4)	0% (0)	100% (10)
Vendors are wearing appropriate uniform	20% (2)	80% (8)	20% (2)	80% (8)	10% (1)	90% (9)
Vendors are wearing gloves	30% (3)	70% (7)	30% (3)	70% (7)	10% (1)	90% (9)
Vendors are wearing hair net	0% (0)	100% (10)	0% (0)	100% (10)	10% (1)	90% (9)
B. Personal hygiene						
Free handlers free of exposed cuts	90% (9)	10% (1)	90% (9)	10% (1)	100% (10)	0% (0)
Fingernails are clean & short	70% (7)	30% (3)	80% (8)	20% (2)	70% (7)	30% (3)

Wash hands before & money transactions	0% (0) 100% (10)	0% (0) 100% (10)	0% (0) 100% (10)
Hands are washed properly, frequently at appropriate time	40% (4) 60% (6)	0% (0) 100% (10)	0% (0) 100% (10)
C. Most important attribute on the raw product			
Appearance	60% (6) Good / 30% (3) Bad	80% (8) Good/ 20% (2) Bad	70(7)Good/30%(3) Bad
Price	Not the same	Same	Not the same
Expiry date	NA	NA	NA
Overall quality	NA	NA	NA
D. Leftover storage			
Back to supplier	0% (0)	0% (0)	0% (0)
In the carts	40% (4)	100% (10)	100% (10)
At the vendor's home	60% (6)	0% (0)	0% (0)
E. Source of raw materials to cook			
Home	0% (0)	0% (0)	0% (0)
Industrial	100% (10)	100% (10)	100% (10)

NA: Not Applicable

Table 3: Microbiological quality of orange juice, cheese caek and meat shawarma samples according to Lebanese Standards Institution (LIBNOR) guidelines of ready to eat food

Microorganism	Lebanese Standards Institution (LIBNOR) guidelines	Microbiological quality of food categories		
	Limit cfu/g	Orange juice (n=10)	Cheese caek (n=10)	Shawarma (n=10)
Total count	<10 ⁵ (cheese caek) <10 ⁷ (shawarma)	A	10	10
Yeast	<10 ² (juice)	0	NA	NA
Mold	<10 ² (juice)	1	NA	NA
<i>Staphylococcus aureus</i>	<100 (cheese caek and shawarma)	NA	4	6
<i>L. monocytogenes</i>	ND (cheese caek, shawarma and juice)	10	0	10
<i>Salmonella</i>	ND (cheese caek, shawarma and juice)	10	10	10
Enterohemorrhagic <i>E. coli</i>	ND (cheese caek, shawarma and juice)	10	10	10
Beta glucuronidase <i>E. coli</i>	<100 (cheese caek, shawarma and juice)	10	9	9
Enterobacteriaceae	<10 ⁴ (cheese caek and shawarma)	10	10	10
<i>Clostridium perfringens</i>	<100 (cheese cake and shawarma)	10	10	9

NA: Not Applicable

ND: Not Detectable

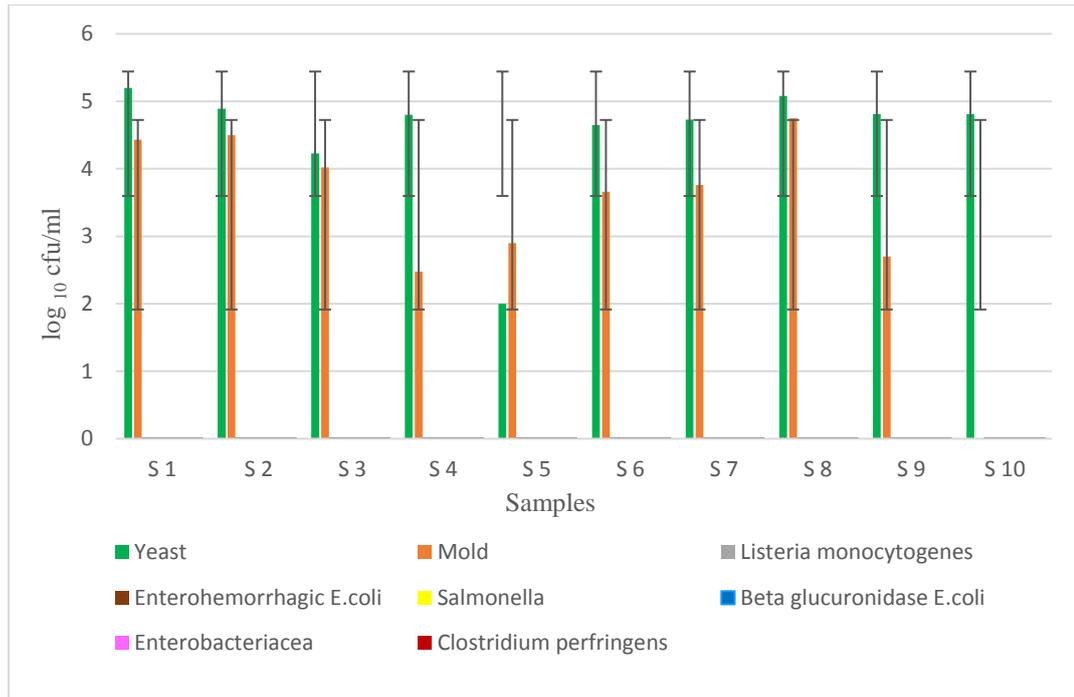


Fig.1: Prevalence of foodborne pathogens, yeast and mold in orange juice samples.

Concerning the total viable count, all the tested samples of RTE food were below the limit recommended by the Lebanese Standards Institution (LIBNOR) (Table 3), where the highest recorded value was 3.85 log₁₀cfu/g (Figure 2,3), which is less than of 7.8 log₁₀cfu/ml and 6.8 log₁₀cfu/ml recorded in similar studies conducted in India and Ethiopia respectively (Kharel, Palni, & Tamang, 2016). No significant difference was found within the total viable count between cheese caek and shawarma samples with a mean value of 3.43 log₁₀ cfu/ g and 3.48 log₁₀ cfu/g respectively (Figure 4). Eromo et al., stated that a high total viable count in foodstuff indicates its poor quality due to cross-contamination and malpractices during food handling (Eromo et al., 2016). In the present study (Table 3), the highest level of *S. aureus* contamination in RTE (4.38 log₁₀ cfu/g) was recorded in S19 (Figure 2,3), 55% of the RTE samples were contaminated with *S. aureus*, these findings were higher than recorded in similar studies conducted in Ghana and Thailand that showed that 39.1% and 17.9% respectively of samples were tested positive for *S. aureus* (Eromo et al., 2016; Ferreira et al., 2016), but less than in India and Benin with 94% and 56.2% of contaminated samples, respectively (Eromo et al., 2016).

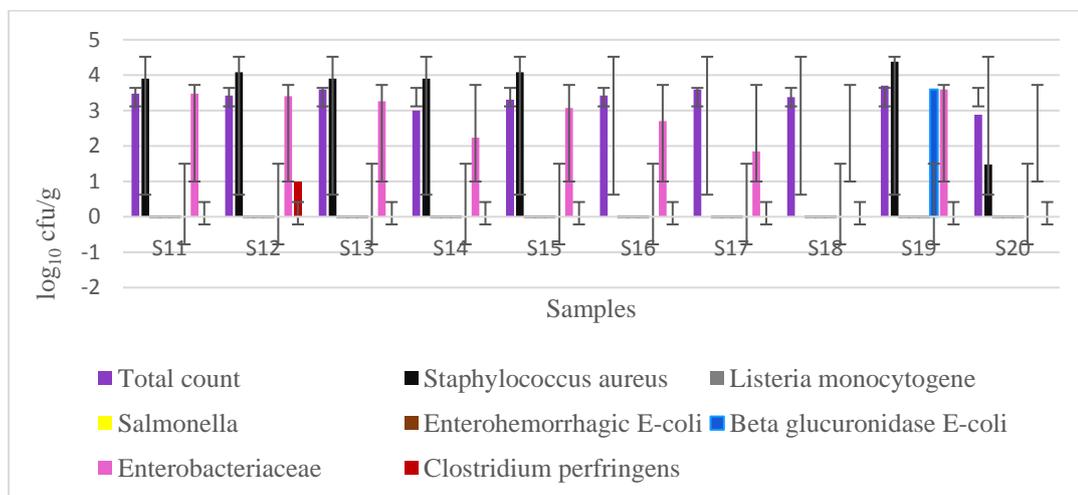


Fig.2: Prevalence of foodborne pathogens in cheese caek samples.

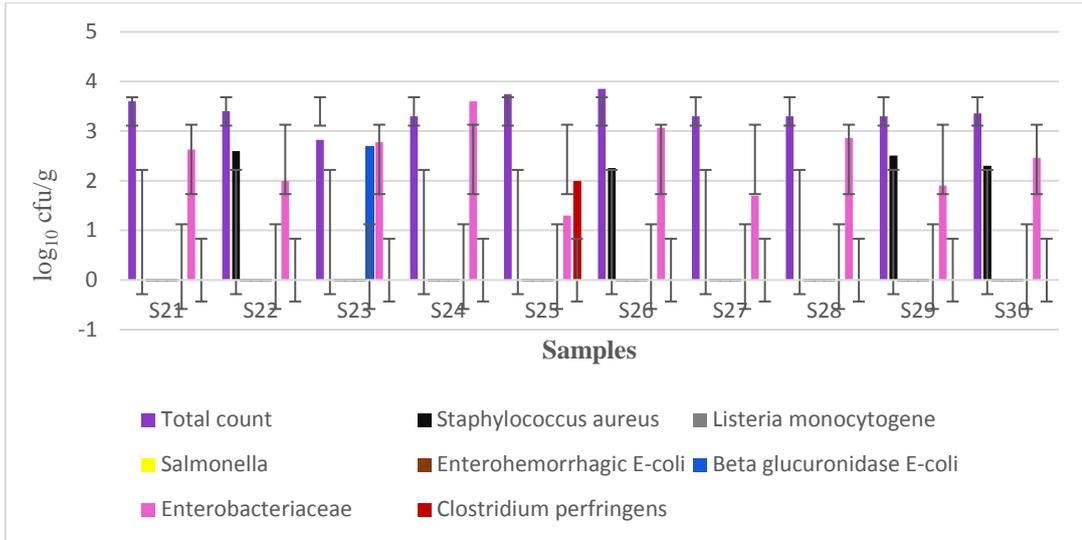


Fig.3: Prevalence of foodborne pathogens in meat shawarma samples.

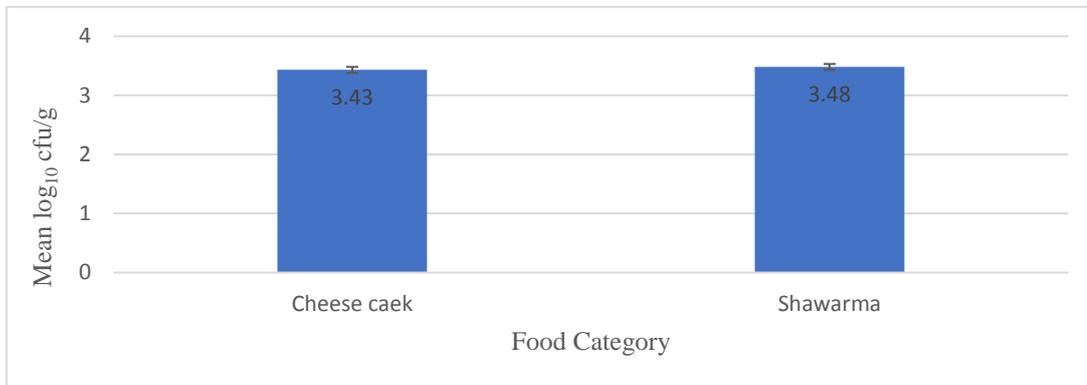


Fig.4: Mean comparison between cheese caek and meat shawarma samples within total viable count (p < 0.05)

The mean comparison between cheese caek and meat shawarma samples within *S. aureus* showed a significant difference between the two food categories, where cheese caek samples mean count (3.96 log₁₀ cfu/g) was higher than shawarma samples (Figure 5). On the other hand, Kharel et al., 2016 stated that the contamination of food samples with *S. aureus* was correlated with the malpractices of street vendors who did not washed their hands frequently, especially after money transaction.

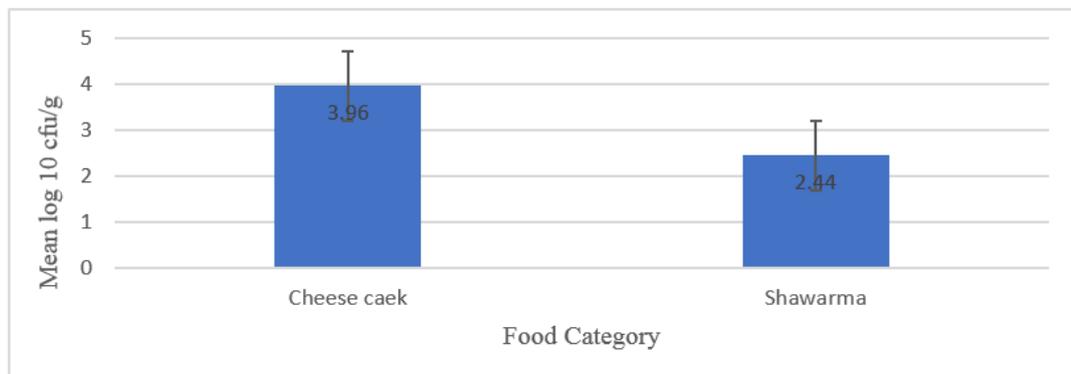


Fig.5: Mean comparison between cheese caek and meat shawarma samples within *S. aureus* (p < 0.05).

This study showed that the highest recorded Enterobacteriaceae load was $3.6 \log_{10}$ cfu/g in S19 (Figure 2,3) less than the standard limit ($4 \log_{10}$ cfu/g) (Table 3), these findings were less than recorded load in Ethiopia with $4.83 \log_{10}$ cfu/g 4 respectively; however, more than recorded value in Egypt with $2.9 \log_{10}$ cfu/g. Mean comparison between cheese caek and shawarma samples within Enterobacteriaceae showed no significant difference between the two food categories (Figure 6). Moreover, the reasons behind a high levels of Enterobacteriaceae in RTE is related to hygienic practices and contamination of processed food (Eromo et al., 2016).

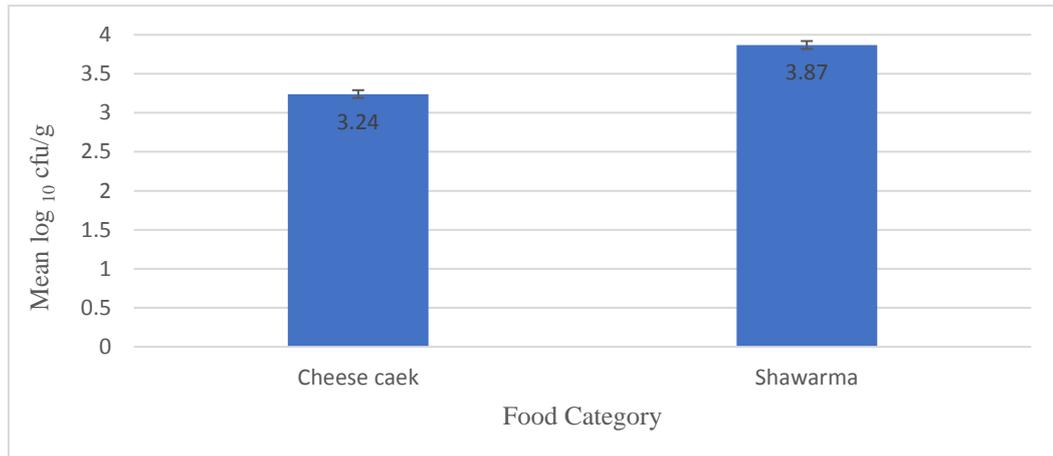


Fig.6: Mean comparison between cheese caek and meat shawarma samples within Enterobacteriaceae ($p < 0.05$).

B-Glucuronidase *E. coli* when detected, was less than $2 \log_{10}$ cfu/g (Table 3), except in one sample of cheese caek ($3.6 \log_{10}$ cfu/g in S19) (Figure 2,3). These findings were less than 20% for a study conducted in Portugal (Campos, Gil, Mourão, Peixe, & Antunes, 2015). Moreover B-Glucuronidase *E. coli* was considered as an indication of fecal contamination (Campos et al., 2015). The reheating of RTF and wrong storage temperature are the main causes of *C. perfringens* in foodstuff. In the present study, *C. perfringens* was detected in two samples one of which did not meet standard and exceeded the limit (Table 3), thus the finding of this study was in accordance with previous study that showed that vegetables, meat and poultry are the most viable source of *C. perfringens*, *L. monocytogenes*, *S. aureus* and *Salmonella* contamination which is the real case of shawarma samples (Rane, 2011). *L. monocytogenes*, *Salmonella* and Enterohemorrhagic *E. coli* were not detected in the studied samples (Table 3).

3.4 Prevalence of *Staphylococcus aureus* in Street Food Vendor's Hand

In the present study, 30% of the tested hand swabs were positive for *S. aureus* swabs (Figure 7), more than previous studies performed in Turkey, Lebanon, Jordan and South-eastern Anatolia that showed 0.77% and 5.6% respectively of positive samples for *S. aureus* (El Darra, Raafat, & El-Ghazzawi, 2019). However, they were less than 36% and 53.2% of positive samples for *S. aureus* in performed study in Zimbabwe, and Kuwait, respectively (S Uzunović et al., 2013; Islam et al., 2019).

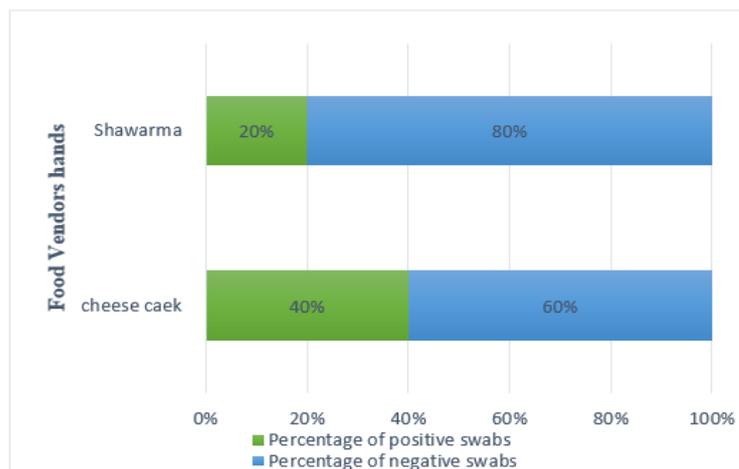


Fig.7: Prevalence of *S. aureus* in hand swabs for cheese caek and meat shawarma vendors in Tripoli.

3.5 Evaluation of the Accordance between Exceeding Standards Levels of *S. Aureus* and Unsatisfactory Food Safety Practices

All wrong practices of food safety spotted in food carts were positively correlated to exceeding limit of *S. aureus* to wrong practices while preparation (Table 4). Although, Islam et al. confirm that up to 70% of food handlers are generally *S. aureus* carrier, assuming that it has a common symptom that people in the third world will not check themselves for it (Islam et al., 2019); a significant correlation was detected between vendor's health status and the occurrence of *S. aureus* in served food. On the other hand, Kharel et al. stated that the vendors who did not washed their hands after money transaction, results in an increase in *S. aureus* level in served food which is considered risky and can cause foodborne disease (Kharel, Palni, & Tamang, 2016); in contrast, no correlation was found between hand washing before and money transaction and *S. aureus* load in street food studied in this research.

Table 4: Correlation between wrong food safety practices and exceeding levels of *S. aureus* in tested RTE samples

Variable	P value (<0.05)
Connectivity to water supply	0.009
Clean, maintained and protected equipment and utensils	0.009
Suitable storage of raw material	0.034
Food Packaging meeting Standard	0.009
Immediate disposal of leftover	0.009
Vendors appears in good health	0.017

3.6 Antibiotic Susceptibility of *S. Aureus*

Table 5 showed that There is only one MRSA in vendor's hand (H29) with inhibition zone =20 mm against cefoxitin, MRSA have been considered as one of the major causes of foodborne diseases globally, since it can acquire resistance rapidly over many antibiotics, rendering it a life threatening strain (Akanbi et al., 2017; Islam et al., 2019). Moreover, around 50-70% of the healthy vendors are carriers of *S. aureus* leading to food cross-contamination through wrong hygienic practices (Islam et al., 2019). The rest 3 isolates (cheese caek against tetracycline, cheese caek and shawarma against oxacillin) considered as MDR.

Table 5: Antimicrobial resistance pattern of *S. aureus* isolated from cheese caek, shawarma samples and vendor's Hands

Antibiotic	Disc content (µg)	Resistant Breakpoint (zone of diameter in mm)	Source	No. (%) of resistant <i>S. aureus</i> isolates			
				Cheese caek (n=7)	Shawarma (n=4)	Vendor's hands (n=6)	Total (n=17)
Tetracycline (T)	30	≤ 14	CLSI 2020	1 (14.3)	0 (0)	0 (0)	1(5.88)
Amoxicillin (A)	25	≤ 14	CA-SFM (1996)	0 (0)	0 (0)	0 (0)	0 (0)
Oxacillin (OX)	1	≤ 10	CLSI 2020	1 (14.3)	1 (25)	0 (0)	2 (11.76)
Cefoxitin (FOX)	30	≤ 21	CLSI 2020	0 (0)	0 (0)	1 (16.7)	1(5.88)

4. CONCLUSION

Globally, Foodborne illnesses are one of the most common public health problems. Studies have shown that RTE foods served by street carts act as vehicles of foodborne pathogens from vendors to consumers. To our knowledge, this is the first study to report the effect of hygiene practices on microbiological contamination of food carts in Tripoli, North Lebanon. The unsatisfactory level of foodborne pathogens detected in food samples, which is caused by inadequate hygiene practices, poses serious health risk to the population with weakened immune system. Therefore, it is crucial to take actions at the level of ministry of public health where relevant food safety legislations should be set and applied with frequent monitoring to make sure that end consumer is receiving safe RTE food.

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