International Food Research Journal 20(5): 2973-2977 (2013)
Journal homepage: *http://www.ifrj.upm.edu.my*

Short communication

Microbiological quality on food handlers' hands at primary schools in Hulu Langat District, Malaysia

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Article history

Received:
Received in revised form:

Accepted:

Keywords

Cross-contamination
Fecal contamination
Microbiological quality
Personal hygiene
School foodservice operation

Abstract

A total of 85 food handlers participated in this study to determine the hygienic status of their hands in primary schools located in the state of Selangor (Malaysia). Overall findings revealed that the fecal contamination and personal hygiene of the food handlers were well maintained with the range of mean bacterial counts from 0.18 to 0.47 $\log_{10}$ Colony Forming Units/cm^2 during the three intervals of hand swabbing (before, during and after) preparation of ready-to-eat foods. However, the general indication of the microbiological quality (Aerobic Plate Count) was out of the standard (range of mean bacterial counts from 1.39 to 1.56 $\log_{10}$ Colony Forming Units/cm^2) based on previous literature. This study highlighted that the food handler's adherence to Good Manufacturing Practice and Sanitation Standard Operating Procedures was insufficient and suggested that attention should be emphasized on their practices at the intervals of school recess: before, during and after the preparation of ready-to-eat foods. In addition, there is also a need in the implementation of an effective HACCP program in Malaysia school foodservice operations.

Introduction

Microbiological hazards are considered to be a great challenge to food safety due to the potentially harmful microorganisms that have the capability to multiply rapidly from extremely small amount in food or in the human body after consumption (Tent, 1999). Unfortunately, according to local news Boey (2010), most food poisoning cases in Malaysia are caused by eating food contaminated with infectious agents where the food contains harmful viruses, bacteria and parasites. *Escherichia coli* (*E. coli*), *Salmonella*, *Shigella* and *Staphylococcus* are the common infectious bacteria which cause food poisoning as these bacteria will release poison that causes gastroenteritis. In addition to that, multiple surveillance on the contamination of *Bacillus cereus* Lee *et al*. (2009) in ready-to-eat cereals, *Campylobacter* spp. in vegetables Chai *et al*. (2007) and etc, showed the prevailing contamination in foods and the role in of food handler's hygiene practices in the increment or reduction of the level of contamination. As a result, preventive measures should be taken to ensure that the outbreaks of foodborne illnesses can be minimized.

Human hands may be the single most important of all disease transmissions. Human hands are in regular contact with the surrounding environment and a variety of pathogens can reach the mucous membranes in the mouth, nose, eyes and genitals of human being through the hands and consequently contribute to foodborne illness outbreaks Hawker *et al*. (2012). Food can become contaminated via dirty hands if there is a lack of proper hand hygiene among the food handlers when handling food.

Proper hand hygiene is needed among workers in foodservice operations as hands had been indicated as a potential vehicle for transferring food poisoning bacteria (Guzewich and Ross, 1999; Gorman *et al.*, 2002; Dharod *et al*., 2009). Poor hand hygiene may contribute to high levels of *S. aureus* and *E. coli* on the hands of food handlers as verified in the finding of (Ayçiçek *et al*., 2004). Hence, good hand hygiene practices should be practiced all the time regardless if hands are dirty or clean, as Staskel *et al.* (2007) concluded that even if a surface seems clean, it may already be harbouring microorganisms. Limited data was available for local studies on the microbiological quality of food handlers' hands especially in primary schools which involves the vulnerable young

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children. Thus, this study was carried out to examine the microbiological quality of food handlers' hands by using Aerobic Plate Count (APC), *E. coli*/Coliform and *S. aureus* counts.

Materials and Methods

Samples collection

A total of 1020 samples were collected from 85 food handlers' hands at 38 primary schools in Hulu Langat district. Microbiological analysis was conducted on food handlers' hands to test for APC, *E. coli*/Coliform and *S. aureus* counts. Duplicate samples were collected from the palm of each hand of the food handlers. The collection was done during weekdays (06:00-10:00) at the following intervals; before, during and after preparation of ready-to-eat (RTE) foods such as 'nasi lemak', sandwiches, fried foods and burgers.

Sterile swabs (Premier, China) were removed from coded test tubes that contained 5 ml of 1x sterile phosphate-buffered saline (Oxoid, Basingstoke, UK) and the targeted areas (palms of food handlers) were swabbed. Sampling was performed by swabbing the areas horizontally, vertically and diagonally by using aluminium templates. The procedures were done aseptically to minimize the risk of contamination. Swabs were then placed back into the coded test tubes. The collected samples were stored and transported in insulated boxes filled with crushed ice prior to analysis. The storage temperature was within 0-4ºC while the transport duration to the laboratory was within 15 min to one h. Analyses were performed immediately upon arrival to the laboratory.

APC, E. coli/Coliform and S. aureus counts on food handlers' hands

Each of the food handler's palms were analyzed for APC, *E. coli*/Coliform and *S. aureus* counts using Petrifilm Aerobic Count Plates, Petrifilm *E. coli*/Coliform and Petrifilm Staph Express Count Plates respectively (3M Microbiology, St. Paul, USA). Swab contact method on Petrifilm plates was used to evaluate APC, *E. coli*/Coliform and *S. aureus* counts on food handlers' palms (George *et al*., 2001). Upon arrival at the laboratory, the test tubes and the swabs were shook vigorously for 10 sec to release bacteria from the swabs. Next, the swabs were discarded and 1 ml of the samples was poured onto respective Petrifilm plates. The top film was then rolled down onto the sample. Subsequently, the recessed side of the plastic spreader plate was placed on the center of the Petrifilm plate and was gently pressed to distribute the sample evenly. The spreader was removed and the Petrifilm plate was left for 1 min to permit the gel to form.

Petrifilm plates were then incubated in a horizontal position with the clear side up in stacks of no more than 20 plates at 30 ± 1ºC for 48 h ± 2 h for APC and 35 ± 1ºC for 24 ± 2 h for *E. coli*/Coliform and *S. aureus* counts. After incubation, the plates were counted on a standard colony counter as per manufacturer instructions and the results were recorded as number of colony forming units (CFU) per 10 cm^2. For colony enumeration, all red colonies regardless of sizes and intensities that appeared on Petrifilm APC plates were counted. For *E. coli*/Coliform counts, all blue colonies associated with gas appeared on the plates were counted as *E. coli* whereas all red colonies associated with gas appeared were counted as coliforms colonies. All red-violet colonies on Petrifilm Staph Express Count Plates were counted as *S. aureus* whilst black or blue-green colonies were further confirmed by using Petrifilm Staph Express Disk. The number of Colony Forming Units was reported as $\log_{10}$ CFU per cm^2 as uniform samples could be obtained.

Statistical analysis

Statistical analyses were performed using SPSS Statistics 19. Statistical significance for all tests was set at the level of $p \leq 0.05$ and descriptive statistics were calculated for all variables as appropriate. Mean bacterial counts on APC, *E. coli*/Coliform and *S. aureus* for the intervals before, during and after RTE foods preparation were compared using nonparametric tests whereas the intervals of RTE foods preparation for the three indicators were compared using both one-way analysis of variance (one-way ANOVA) and nonparametric tests.

Results and Discussion

Microbiological quality on food handlers' hands

Table 1 reported the mean of APC, *E. coli*/Coliform and *S. aureus* counts on 85 food handlers' hands in school food service operations. The data collected were from both right and left palms of the food handlers and there was no significant difference found between them on the mean bacterial counts. Nevertheless, in the study of Ayçiçek *et al.* (2004), it has been noticed that the bacteria isolated from right hands were significantly higher than the left hands of the food handlers from the military training hospital in Turkey as all of them were right-handed.

Table 2 reports the percentage of incidence of APC, *E. coli*/Coliform and *S. aureus* counts on food handlers' hands in school foodservice operations (n = 85). In the present study, there were significant

Table 1 Mean of APC, *E. coli*/Coliform and *S. aureus* counts on food handlers' hands in school foodservice operations (n = 85)

Intervals	Bacterial Count[a]		
	Aerobic Plate Count	*Escherichia coli*/Coliform Plate Count	*Staphylococcus aureus* Plate Count
	Mean ± Standard deviation	Mean ± Standard deviation	Mean ± Standard deviation
Before	$1.56^{aA} \pm 0.58$	$0.22^{bA} \pm 0.39$	$0.47^{cA} \pm 0.67$
During	$1.41^{aA} \pm 0.55$	$0.18^{bA} \pm 0.36$	$0.31^{bA} \pm 0.53$
After	$1.39^{aA} \pm 0.63$	$0.24^{bA} \pm 0.41$	$0.43^{bA} \pm 0.66$

[a] Data were reported as mean ± standard deviation for four replicates ($\log_{10}$ Colony Forming Units/cm^2).
* Different lowercase superscript letters within the row differ significantly ($p < 0.05$).
* Different uppercase superscript letters within the column differ significantly when ($p < 0.05$).

Table 2 Percentage of incidence of APC, *E. coli*/Coliform and *S. aureus* counts on food handlers' hands in school foodservice operations (n = 85)

Intervals	Positive strains					
	Aerobic Plate Count		*Escherichia coli*/Coliform Plate Count		*Staphylococcus aureus* Plate Count	
	n	%	n	%	n	%
Before	85	100.00	61	71.76	63	74.12
During	85	100.00	61	71.76	56	65.88
After	85	100.00	58	68.24	60	70.59

differences (χ^2 (2) = 129.071, p = 0.000) on the mean bacterial counts before RTE foods preparation for APC, *E. coli*/Coliform and *S. aureus* counts. The mean bacterial counts for APC (M ± SD = 1.56 ± 0.58) were the highest followed by S. aureus counts (M ± SD = 0.47 ± 0.67) and eventually *E. coli*/Coliform counts (M ± SD = 0.22 ± 0.39). Similarly, there were also significant differences on the mean bacterial counts during and after RTE foods preparation (χ^2 (2) = 137.162, p = 0.000 and χ^2 (2) = 107.801, p = 0.000) for the three indicators stated. The mean bacterial counts for APC were higher than *S. aureus* and *E. coli*/Coliform counts. However, there was no significant difference on the intervals of RTE foods preparation for the three indicators tested. As Staskel *et al.* (2007) had pointed out that the limitation in their study on microbiological evaluation of foodservice surfaces was the time of sample collection; this study may serve as a reference as it proved that the sample collection time did not influence the mean bacterial counts on food handlers' hands.

The standard of bacterial counts were based on the study of Sneed *et al.* (2004) who stated that less than 1.3 $\log_{10}$ CFU was used for APC, less than 1.0 $\log_{10}$ CFU for Enterobacteriaceae counts, and less than 1.0 $\log_{10}$ CFU for *S. aureus* counts were considered 'acceptable'. APC for the three stages were out of the standard set by Sneed *et al.* (2004) whereas *E. coli*/Coliform while *S. aureus* counts were within the standard set. Although the counts for *E. coli*/Coliform and *S. aureus* were acceptable, food handlers from the primary schools in Hulu Langat district should reduce the counts for these pathogenic bacteria to the minimum value as these bacteria may bring detrimental health effects to the school children which are the vulnerable group.

For the percentage of incidence of APC, *E. coli*/Coliform and *S. aureus* counts on food handlers' hands, the results were reported in Table 3. APC was found to be the highest among the other two indicators as all of the food handlers' hands contained aerobic bacteria. For *E. coli*/Coliform, the range of incidence was from 68.24% to 71.76% while the range of incidence for *S. aureus* was from 65.88% to 74.12%. In a retail group in South Africa, it has also been reported that the presence of total viable counts on food handlers' hands was the highest (98%) compared to the coliform (40%) and *S. aureus* (88%) counts (Lues and Van Tonder, 2007). Food handlers should improve on good hand hygiene practices as the incidence of APC on food handlers' hands was extremely high. Good Manufacturing Practice (GMP) and Sanitation Standard Operating Procedures (SSOP) are the two mandatory aspects that every food handlers should comply with to ensure the safety of the food produced. Attention should also be focused on *S. aureus* isolates as recently, a food worker's hand had been contaminated by toxic shock syndrome-1 (TSST-1) secreted by *S. aureus* in a restaurant in Spain (Sospedra *et al.*, 2012). TSST-1 can cause symptoms which include high fever, headache, confusion, vomiting, diarrhoea and acute renal failure in children.

Table 3 below displays the standard of food handlers with hands swab samples results. According to the standard given by Sneed *et al.* (2004), the percentage of food handlers that comply with the mean bacterial counts for APC was very low compared to the percentage of food handlers who did not meet the mean bacterial counts. These results

Table 3 Percentage of 85 food handlers with hands swab samples results within and out of standard (as set in Sneed *et al.* (2004) of bacteria counts

		n (%)		
		Aerobic Plate Count	*Escherichia coli*/Coliform Plate Count	*Staphylococcus aureus* Plate Count
Before	M	20 (23.53)	79 (92.94)	65 (76.47)
	O	65 (76.47)	6 (7.06)	20 (23.53)
During	M	28 (32.94)	77 (90.59)	73 (85.88)
	O	57 (67.06)	8 (9.41)	12 (14.12)
After	M	29 (34.12)	76 (89.41)	65 (76.47)
	O	56 (65.88)	9 (10.59)	20 (23.53)

Note: Acceptable levels are expressed as the number of viable bacteria expressed as $\log_{10}$ CFU/cm^2; M = meeting the standard, O = out of standard.

demonstrate that effective Hazard Analysis and Critical Control Points (HACCP) program was not applied in the primary schools in Hulu Langat district as Hong *et al.* (2008) had stated that APC was a useful tool in evaluating the satisfactory of HACCP plan. Food handlers that comply with the mean bacterial counts for APC before, during and after preparation of RTE foods were only 23.5%, 32.9% and 34.1%. On the contrary, percentage of food handlers that comply with the mean bacterial counts for *E. coli*/Coliform and *S. aureus* were quite high compared to the percentage of food handlers that exceeded the mean bacterial counts. Food handlers that comply with the mean bacterial counts for *E. coli*/Coliform and *S. aureus* before, during and after preparation of RTE foods were 92.4%, 90.6%, 89.4% and 76.5%, 85.9%, 76.5% respectively. Balzaretti and Marzano (2013) had also stated that the food handlers in northern airport Italy restaurants were examined on TPC and coagulase positive *staphylococci* counts within the satisfactory limit (91.6% and 96.5% respectively).

Conclusions

In general, the fecal contamination and personal hygiene of the food handlers from primary schools in Hulu Langat district were well maintained. Maintaining good hand hygiene is important as pathogenic bacteria such as *E. coli* and *S. aureus* may bring harmful health effects to the school children as they are considered as a high-risk group. For the general indication of microbiological quality, food handlers should adhere to GMP and SSOP as the APC investigated before, during or after the preparation of RTE foods was not satisfactory.

Acknowledgements

The authors would like to express their sincere thanks to all of the principles of the respective schools and also the owners of the school canteens for allowing them to carry out sample collection at the canteens. Besides, the authors also like to express gratitude to Universiti Putra Malaysia for the Research University Grant Scheme.

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