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Occurrence of Foodborne Pathogens in Chickens Sandwiches Distributed in Different Supermarkets of Tehran Province, Iran

Zohreh Mashak [1]; Hamidreza Sodagari [1,*]; Behrooz Moraadi [2]; Ashkan Ilkhanipour [1]

[1]Department of Food Hygiene, College of Veterinary Medicine, Karaj Branch of Islamic Azad University, Karaj, IR Iran
[2]Department of Microbiology, College of Science, Karaj Branch of Islamic Azad University, Karaj, IR Iran

*Corresponding author: Hamidreza Sodagari, Department of Food Hygiene, College of Veterinary Medicine, Karaj Branch of Islamic Azad University, Karaj, IR Iran, P.O.Box:31485313. Tel: +98-9122186315, E-mail: hr_sodagari@yahoo.com

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Background: Increasing urbanization, immigration and tourism has changed the human lifestyle. This modern lifestyle has demanded safety, quality, and fast availability of ready to eat (RTE) foods like chicken sandwiches.
Objectives: For presentation of proper solutions regarding food safety, identification of pathogens in different foods is necessary. Therefore, the present study was carried out to assess the microbiological quality of chicken sandwiches distributed in Tehran province, Iran.
Materials and Methods: A total of 200 chicken sandwich samples (chicken sausage, chicken fillet, minced chicken fillet) were purchased from different supermarkets in Tehran city randomly during 2013 and transported to the laboratory of food hygiene of Islamic Azad University, Karaj branch under temperature-controlled conditions for bacteriological examination by American Public Health Association (APHA) method.
Results: The average count ± standard error (and percent of unacceptable samples) of *S. aureus*, *B. cereus* and *Coliform* were 1.6 ± 0.56 (28%), 2.0 ± 0.62 (10%), 4.2 ± 1.12 (50%) CFU/g, respectively. Moreover, *E. coli* and *Salmonella* spp. were identified in 21% of chicken sandwich samples.
Conclusions: The large number of foodborne pathogens detected in this study, represented a potential health hazard to consumers. Thus, it is necessary to employ Good Hygiene Practices (GHP) and Hazard Analysis and Critical Control Points (HACCP) in order to minimize the risk caused by secondary contamination.

*Keywords:*Chicken; Foodborne Diseases; Iran

1. Background

Increasing urbanization, immigration, tourism and world food trade has changed the lifestyle, socio-economic status, cultural behavior and pattern of the food system chain. This modern lifestyle made the consumers to demand the safe, quality and fast available ready to eat (RTE) foods like chicken sandwiches.

Outbreaks of foodborne diseases after eating RTE foods have been reported worldwide (1). One of these outbreaks occurred in the United States at an incidence of 17.6 illnesses per 100000 persons, 2290 hospitalizations, and 29 deaths in 2010 (2). Another investigation showed that, after cooking ingredients, foodborne pathogens in sandwiches may proliferate due to low numbers of other microorganisms (3). Whereas RTE foods do not need any heating prior to consumption, they are categorized as high-risk foods.

One of the most common RTE foods in Iran is the chicken sandwich that includes chicken sausage, cooked chicken fillet with tomato, lettuce, spices, herbs and other things as ingredients. These foods are often prepared by hand, and this direct contact may lead to the increase the incidence of contamination with potential foodborne pathogens (4).

2. Objectives

Despite significant progress in food safety management, occurrence of foodborne disease remains a major problem in food industry. For presentation of proper solutions in the food safety, identification of pathogens is necessary. Thus, the present study was carried out to assess the microbiological quality of chicken sandwiches distributed in Tehran province, Iran.

3. Materials and Methods

3.1. Sample Collection

A total of 200 chicken sandwich samples (chicken sausage, chicken fillet, minced chicken fillet) were bought from different supermarkets in Tehran city randomly during 2013. All packed sandwiches were transported to the laboratory of food hygiene in Islamic Azad University, Karaj branch, under temperature-controlled conditions for bacteriological examination. Bacterial tests followed American Public Health Association instructions (5).

Table 1. Results of Microbial Contamination of Chicken Sandwiches (CFU/g)

Microbial Organism	Min-Max	Mean ± SE	Unacceptable Samples, No. (%)	Sample Size, No.
S. aureus	0.6-6.4	1.6 ± 0.56	56 (28)	200
B. cereus	0.7-5.6	2.0 ± 0.62	20 (10)	200
Coliform	0.9-5.9	4.2 ± 1.12	100 (50)	200
E. coli			42 (21)	200
***Salmonella* spp.**			42 (21)	200

3.2. Sample Preparation

Samples were opened aseptically, and 25 g of each sample was transferred into 225 mL of sterile buffered peptone water in stomachers bag and homogenized. Double culturing was done, after preparation of 10 fold serial dilutions.

3.3. Identification and Numeration of S. aureus

Identification and numeration of *S. aureus*, were done as follows: enrichment of 1 g sample in 10 mL cooked meat medium (Difco), streaking a loopful of the 24-hour enrichment culture on Baird–Parker agar (BPA, Merck) containing egg yolk and potassium tellurite (Merck), and finally, incubation at 37°C for 48 hours.

3.4. Identification and Numeration of Coliform and E. coli

Coliform and *E. coli* were counted by pour plate method in Violet Red Bile agar (VRBA, Merck) with incubation at 37°C for 48 hours. After transferring suspected colonies to Brilliant Green Bile Lactose Broth, incubation was done at 37°C and 44°C for 48 hours respectively. For detecting *E. coli*, Eosin methylene-blue lactose sucrose agar (EMBA, Merck) was used and subjected to biochemical tests.

3.5. Identification and Numeration of Bacillus cereus

Surface plate method on *Bacillus cereus* selective agar (Merck) were used for identification of typical *B. cereus* colonies and incubated at 37°C for 24 hours.

3.6. Identification of Salmonella

For identification of *salmonella* spp, 25 g of each food sample was pre-enriched in lactose broth (Merck) at 37°C for 18 hours. Then, 1 mL was transferred into 10 mL selenite cystine broth (Merck) for enrichment, incubated at 37°C for 24 hours. Finally, we used *Salmonella Shigella* (SS) agar (Merck), bismuth sulfite agar (Merck) as selective media, triple sugar iron agar (Merck), lysine iron agar (Merck) as differential media and Urease (Merck) as complement media.

4. Results

Table 1 presents microbial analyses of chicken sandwich samples. The average count ± standard deviation (and percentage of unacceptable samples) of *S. aureus*, *B. cereus* and Coliform were 1.6 ± 0.56 (28%), 2.0 ± 0.62 (10%), and 4.2 ± 1.12 (50%) CFU/g , respectively (Table 1)(6). Moreover, *E. coli* and *Salmonella* spp. were identified in 21% of chicken sandwich samples. (Table 1)(6).

5. Discussion

Nowadays, due to lifestyle changes, most of the people do not have enough time to cook, and that is the main reason for increasing RTE foods consumers. However, the safety of these foods should be the first priority since they do not receive any heat treatment before consumption. *S. aureus* is one of the main causes of outbreaks of foodborne diseases in the world that is transmitted by food handlers' skin, nose or hand.

In this study, *S. aureus* range and mean ± SE were found 0.6-6.4 and 1.6 ± 0.56 CFU/g respectively. This pathogen was isolated from 28% of chicken sandwich samples that is hazardous for consumers' health. This finding is higher than previous results that were obtained in Kerman city (4.5%) and Korea (1.3%) (7, 8) from sandwiches.

Many researchers have reported that *B. cereus* is the pathogen that contaminates both raw (9) and processed meat products (10), because this pathogen is ubiquitous, spore forms and highly resistant to adverse conditions such as heat and dehydration (11). In our research, the range and mean ± SE of *B. cereus* were 0.7-5.6 and 2.0 ± 0.62 CFU/g respectively. In the present study, *B. cereus* was detected unacceptable in 20 (10%) of 200 examined samples, which were less than Umoh and Odoba (12) survey on street food (26.3%) but similar to Jang et al. (8) investigation on sandwich.

Coliform bacteria are a common bacterial indicator of sanitary quality of foods. In the present study, 100 (50%) out of 200 examined samples were contaminated to this indicator organism. This rate was lower than finding of Jang et al. (80.9%) (8). *E. coli* is an important fecal indicator that belongs to *Enterobacteriaceae* family like Salmonella. In this survey *Salmonella* spp. similar to *E. coli* were detected unacceptable in 42 out of 200 (21%) chicken sandwich samples. These results are an indicator of poor hygiene practices during preparation, or improper storage (13).

Previous study in Kerman showed higher results for *E. coli* (40.3%) and lower for *Salmonella* (3%) compare to our findings (7). Using same knife, board and gloves without wash-

ing hands, to cut up raw and cooked chicken meat, are the most important causes of cross-contamination.

Several factors may contribute to the presence of pathogens in RTE foods, including poor handling practices and processing, high storage temperatures and cross contamination from food contact surfaces (14). Chicken meat contamination with pathogens is one of the most important reasons of chicken sandwich contamination. In a survey in Turkey, *Salmonella* spp. and *E. coli* O157: H7 were isolated in 18.4% and 4.8% of chicken carcasses respectively (15).

Furthermore, the initial microbiological load on the ingredients of these foods is important too. As wide ranges of vegetables such as tomato and lettuce, spices, herbs, sauce, and so on are used as ingredients in chicken sandwiches. Contamination of these ingredients by enteric pathogen bacteria happen because of many reasons like growing areas with poor water and fertilizer quality. This issue is of serious concern, because most of these ingredients specially vegetables are consumed without major processing.

Several studies were done regarding the microbiological quality of various RTE food ingredients. In 2006, multistate outbreaks of *E. coli* O157: H7 infections through consumption of contaminated lettuce resulted in 71 illnesses in 5 states (16). In the study of Moreira et al. (17), on microbial contamination of various commodities such as spices, 13 (5.6%) of 233 samples were positive for Salmonella.

The large number of indicator and pathogen microorganisms were detected in chicken sandwich samples of the present study represented the potential health hazard to consumers. Therefore, GHP and HACCP should be used on the control of cross-contamination during preparing these foods for reducing foodborne pathogens.

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