A survey on the prevalence of *Salmonella* and coliforms in unpasteurized Iranian cheese using conventional culture method

Jafar Akbarmehr^1^* and Jalil Khandaghi^2^

^1^Department of Microbiology, Islamic Azad University Sarab Branch, Sarab, Iran.
^2^Department of food science, Islamic Azad University Sarab Branch, Sarab, Iran.

Accepted 31 October, 2011

The aim of this study is to evaluate the hygienic quality of unpasteurized cheese by microbiological tests and show the possible contamination of them with coliforms and *Salmonella* spp. A total of 200 samples of unpasteurized cheese were collected randomly from different markets in Sarab city (East Azarbayjan province, Iran) from February 2011 till August 2011. Based on the results of bacteriological tests all of the examined cheese samples showed different degree of coliform contamination. Coliform counts were varied from $1.1 \times 10^2$ – $4.3 \times 10^5$ CFU/g. Out of 200 examined cheese samples 10 (5%) had less than $10^2$ CFU/g, 20 (10%) had more than $10^4$ CFU/g and the other 170 (85%) ranged from $10^2$ - $10^4$ CFU/g. Also 84 (42%) of contaminated cheese samples had positive results for faecal coliform bacteria. The results of bacteriological tests indicated that all the examined cheese samples were absent from *Salmonella* spp. This study revealed the low hygienic quality of local cheese produced in Sarab city. Therefore consumption of this type of cheese may lead to potential hazards for human health which must be considered by health organizations in Iran.

Key words: Cheese, unpasteurized, coliform, sarab.

INTRODUCTION

Milk and other dairy products contain important components of a healthy diet such as: vitamins, vital minerals and high quality proteins but if consumed unpasteurized they also can cause health hazard due to contamination with many kinds of pathogenic bacteria (Lejeune and Rajala-Schults, 2008). These bacteria can gain access to dairy products from infected animals via milk secretions or by secondary contamination occurring during post processing. Coliform bacteria, *Salmonella*, *Mycobacterium bovis*, *Brucella*, and *Listeria* may contaminated the dairy products including cheese which are made from unpasteurized milk or may have been made under unsanitary condition (Doyle and Beuchat., 2007; Jay, 1992). Coliforms are facultatively-anaerobic, Gram-negative, non-sporulating and rod shaped bacteria which include: *Escherichia*, *Enterobacter*, *Klebsiella*, and *Citrobacter*. These micro-organisms are considered as a collection of relatively harmless bacteria that live in large numbers in the intestinal tract of humans and warm-blooded animals. Coliforms are always found in raw milk but with sanitary methods of production number of them can be kept very low. A specific subgroup of coliforms is the faecal coliforms bacteria, the most common member being *E. coli*. These bacteria may be separated from other coliforms by their ability to make infection and are asso-ciated with the faecal material of humans and animals (Marvin, 1976). Coliform bacteria can result from the environment naturally or be faecal in origin. The presence of faecal coliform in food indicates that the food has been contaminated with the faecal materials and is an indicator for a potential health risk (Aman et al., 2009). Although cheese is generally considered to be relatively safe food, if consumed unpasteurized can be health hazard due to contamination with coliforms and numerous of other pathogenic bacteria (Lejeune and Rajala-Schults 2008). *Salmonellosis* is
another food borne infection that may result when unpasteurized dairy products are consumed. *Salmonella* spp are spread with intestinal contents or excrement of infected animals. They also may be spread to dairy products during processing or by improper handling (Doyle and Beuchat, 2007). Unfortunately, despite the considerable attention which has been paid to improve the quality of milk over many years, still the hygienic quality of dairy products are not satisfactory in many under-developed countries. Sarab city is located in a mountainous region in East Azarbayjan province, Iran. The consumption of local and unpasteurized cheese is very popular in this city but until now their contamination with coliforms and *Salmonella* has not been documented. The present study was carried out with an objective to evaluate the hygienic condition of unpasteurized cheese produced in Sarab city by micro-biological tests and show the possible contamination of them with coliforms and *Salmonella* spp.

**MATERIALS AND METHODS**

A total of 200 samples of local cheese (unpasteurized) were collected under sterilized conditions from different markets in Sarab city from February 2011 till August 2011. Each sample was represented by 100 grams of local cheese and were transferred to microbiology laboratory in sterile condition. Bacteriological examinations were carried out according to standard procedure of dairy products (Harrigan, 1998). 10 grams of each cheese sample were added to 90 ml of normal saline. They were mixed properly and then ten-fold dilutions were prepared for each sample. An inoculum of 0.1 ml of each dilution was placed on the surface of MacConkey agar (Merk, Germany) and incubated for 24 h at 37°C. After incubation, plates inoculated with a sample dilution yielding between 30-300 colonies were chosen for coliform count (Quinn et al., 1994). Identification of true faecal type *E. coli* was applied by Eijkman test using Brilliant green bile broth (BGBB) and then confirmed by IMViC tests as described by Quinn et al. (1994). For isolation of *Salmonella* spp the standard bacteriological procedure was used as described by Harrigan (1998).

**RESULTS**

According to the results of bacteriological tests all of the examined cheese samples showed different degree of coliform contamination. Coliform counts were varied from $1.1 \times 10^2$ – $4.3 \times 10^5$ CFU/g. Out of 200 examined cheese samples (Table 2 and Figure 2) 10 (5%) had less than $10^2$ CFU/g, 20 (10%) had more than $10^4$ CFU/g and the other 170 (85%) ranged from $10^2$- $10^4$ CFU/g. Also 84 (42%) of contaminated cheese samples had positive results for faecal coliform bacteria. The results of bacteriological tests for detection of *Salmonella* spp were negative for all examined cheese samples (Figure 1 and Table 1).

**DISCUSSION**

The results given in the Table 1 show that all the coliforms. Also the faecal type *E. coli* was isolated from examined cheese samples were contaminated with 42%
The results illustrated in Table 2 indicate that only 5% of the examined cheese samples had a standard level of coliforms ($10^2 < \text{CFU/g}$) according to Iranian standards (Marhamatizadeh et al., 2005) and the other 95% which ranged from $10^2$ to $10^5 \text{CFU/g}$ had coliforms higher than standard levels. Coliform counts are an indicator for possible faecal contamination and reflect the hygienic standards adopted in dairy processing (Aman et al., 2009; Riemann and Bryan, 1979). The presence of faecal coliforms in food indicates that the food has been contaminated with the faecal materials. E. coli is one of the most important food-borne pathogens and has been implicated in many food-borne outbreaks associated with severe symptoms and highly fatality rate (Doyle and Beuchat, 2007; Brodsky, 1984). Moreover, the occurrence of high proportion of faecal coliforms in cheese samples represents an indication of the presence of other pathogenic bacteria due to low sanitary measures. In our previous work we studied about the prevalence of Brucella in local cheese produced from Sarab city. In this study Brucella spp were found from 2.2% of examined cheese samples (Akbarmehr, 2011). Unfortunately despite the enormous advances in cheese processing technology milk-borne disease outbreaks continue to occur due to consumption of unpasteurized dairy products which can be contaminated with different kinds of pathogenic bacteria. Recent studies in Iran and other countries showed different rates of contamination of cheese with coliforms. A study which was carried out by Mansuri et al. (2006), 77 domestic Iranian soft cheese samples was examined by bacteriological tests. They isolated E. coli from 76 (98.7%) cheese samples indicate a higher contamination rate compared with our results. Marhamatizadeh et al. (2006) studied about the contamination of white traditional cheese with coliform and E. coli in Kazeroun city, Iran. They examined 50 cheese samples and showed coliform contamination in 49 (98%) examined cheese samples which is in agreement with our results in the present study. Many authors from different countries determined the coliform contamination of dairy products (Frank and Marth., 1979; Salman and Hamad, 2011; Aleksieva, 1980).

Nour et al., (2010) compared the microbiological quality of processed and non processed Sudanese white cheese. In this study they concluded that if the Sudanese white cheese could be further processed the hygienic quality would improve. Another study which carried out by Khayat et al. (1987), out of 256 cheese samples which

<table>
<thead>
<tr>
<th>Range of coliform count (Log 10 CFU/g)</th>
<th>Frequency</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No</td>
</tr>
<tr>
<td>&lt;2</td>
<td>10</td>
</tr>
<tr>
<td>2-3</td>
<td>50</td>
</tr>
<tr>
<td>3-4</td>
<td>120</td>
</tr>
<tr>
<td>&gt;4</td>
<td>20</td>
</tr>
<tr>
<td>Total</td>
<td>200</td>
</tr>
</tbody>
</table>

Table 2. Frequency of Coliform bacterial counts in examined cheese samples obtained from Sarab city.
examined for coliforms 46% had less than 100 CFU/g which indicates lower contamination rate compared to the samples which were examined in our study. Isolation of *Salmonella* spp from unpasteurized cheese produced in Sarab city was the second aim of the present work. Fortunately none of the 200 examined cheese samples gave any positive result for presence of *Salmonella* (Table 1 and Figure 1). This result may reveal that *Salmonella* cannot survive after processing or ripening process of cheese. Finally the results of this study revealed the low hygienic quality of local cheese produced in Sarab city. Consumption of them may lead to potential hazards for human health. Therefore we recommend that hygienic measures must be followed during handling, processing and storage of cheese and also pasteurization of milk should be imposed to remove the coliforms and other pathogenic bacteria from dairy products.

**ACKNOWLEDGEMENTS**

This work was supported by the Islamic Azad University of Sarab Branch. We are thankful to Mr. S.S. Taheri for his valuable collaboration.

**REFERENCES**


