Study microbial content of locally produced and sold milk products (the local white soft cheese and the local cream) in Al-Muthanna markets
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ABSTRACT
This study was done to identify some of microbial contaminators for milk products (the soft cheese and the local soft cream) common in Al-Muthanna markets and the evaluation the healthy range from health standardize correspondence. Collecting 50 sample of local soft cheese and 50 sample of local cream during September 2015 to April 2016. Standard bacteriological cultivation and biochemical tests were used to isolate and identify bacterial pathogens that cause microbial contamination in the milk samples. The result showed increase their microbial content which involved many of causing human illness and food poisoning, the most common bacterial strains were isolated from soft cheese and local cream was Staphylococcus aureus (26%, %30) Consequently, It is located within the crisis boundaries to cause food poisoning. The average number of Escherichia coli in soft cheese and local cream was (%20, %24), followed by Pseudomonas aeruginosa, Klebsiella pneumonia and Salmonella sp. (%10, %6) for each genus, Bacillus sp. (16%, 8%), and the results of yeast was 8% for each soft cheese and local cream. The result indicates that strict preventive measures should be adopted to ensure contamination free milk products for the good health of all consumers.

Keywords: microbial content, milk products, white soft cheese, local cream

INTRODUCTION
Milk is supposed to constitute a complex ecosystem for various microorganisms including bacteria. Milk products like cheese and cream are the most demanding foods in all cities of Iraq; there is an increase demand by the consumer for high quality natural food, free from artificial preservatives and contaminating microorganisms (3 & 19).

Production of local soft white cheese and cream is mainly from animal breeders or from small-scale laboratories and sold directly to consumers so it is necessary regard hygienic conditions in production and store (21).

Milk is supposed to constitute a complex ecosystem for various microorganisms including bacteria to suit the moisture and contain many carbohydrate and nitrogen compounds in addition to the availability many vitamins and minerals that make it a habitable environment for micro-organisms that plays an important role in transmission of a number of diseases and a major cause of food poisoning (3, 15 &23).
Contamination of milk and milk products, with pathogenic bacteria is largely due to processing, handling, and unhygienic conditions as well as the presence of intestinal toxins (14& 24).

Local soft cheeses and cream contain various types of microorganisms, aerobic and anaerobic bacterial spores, yeasts and molds in large numbers (18). Due to the high microbial load present in raw milk, pasteurization is commonly used to eliminate all pathogenic and most of nonpathogenic organisms before its further processing into cheese. Moreover, pasteurization of milk for cheese making is intended mainly to reduce microbial load, ensure greater yield, standard level of quality and ripening at higher temperature (7&16).

Although, cheese production should employ the full pasteurization process, there is a long standing tradition of making cheese from raw or heat treated milk. Heat treatment is defined as heating milk at time temperature less rigorous than pasteurization referred as sub-pasteurization. It has no international standards and could destroy most of pathogenic microorganisms with partial inactivation of indigenous enzymes, and other biological components initially present in raw milk (9,20, 22&25).

MATERIALS AND METHODS

Samples collection

20 Sample of cottage cheese and cream were collected during September 2015 to April 2016, from different localities of Al-Muthanna city, and were brought to the laboratories of Al Muthanna university- faculty of science, biology department, for the isolation of the most prominent microorganisms contaminated with local soft cheese and cream.

Samples of each milk product were collected aseptically in sterilized container, then transferred to sterile plastic bags and directly transported to the laboratory under cold conditions, then stored at 4 °C and analyzed within 24 hours.

Microbiological analysis

A Portion (10 g or 10 ml) from the center of each sample was extracted aseptically and homogenized with 90 ml sterile enrichment broth media and incubated at 37 °C for 24 hours, for further biochemical examination.

Media and growth conditions

For the isolation and identification of the most prominent microorganisms contaminated with local soft cheese and cream were determined following aseptic sampling techniques as described in Quinn et al. and Barrow and Feltham (6&17). Briefly: a loop full (0.01ml) of sample in nutrient broth media was streaked on 7% blood agar (Oxoide, Germany) and incubated aerobically at 37°C. The plates were checked for bacterial growth after 24, 48 and 72 hours to rule out slow growing microorganisms and sub-cultured on blood agar at 37°C for 24 hours to get pure culture. A single colony from a pure culture subjected to Grams’ stain to observe morphological characteristics and transferred to MacConkey agar, Manitol Salt Agar, S.S Agar, Levine Eosin Methylene Blue (EMB) Agar and Potato Dextrose Agar (PDA) to be grown for further analysis.

Identification of bacteria to the species level was carried out based on their colony characteristics, Gram’s staining and morphological characteristics, growth on selective media, catalase, and oxidase tests, hydrogen sulfide (H2S) production, indole, methyl red(MR), and Voges-Proskauer (VP) reaction, citrate utilization, oxidation-fermentation test, motility, and different carbohydrate testes (Table 1)
TABLE 1: Biochemical characterization of bacterial isolates.

<table>
<thead>
<tr>
<th>Bacterial properties</th>
<th>Bacterial strains</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S.aureus</td>
</tr>
<tr>
<td>Gram stain</td>
<td>+ ve</td>
</tr>
<tr>
<td>Shape</td>
<td>cocci</td>
</tr>
<tr>
<td>Catalase</td>
<td>+ ve</td>
</tr>
<tr>
<td>Oxidase</td>
<td>- ve</td>
</tr>
<tr>
<td>Motaility</td>
<td>-ve</td>
</tr>
<tr>
<td>Citrate</td>
<td>+ ve</td>
</tr>
<tr>
<td>H2S</td>
<td>-ve</td>
</tr>
<tr>
<td>MR</td>
<td>+ ve</td>
</tr>
<tr>
<td>Indole</td>
<td>-ve</td>
</tr>
<tr>
<td>Nitrate reduction</td>
<td>+ ve</td>
</tr>
<tr>
<td>VP</td>
<td>+ ve</td>
</tr>
</tbody>
</table>

RESULTS AND DISCUSSION

It appears from the results recorded in table (2) that the average number of bacteria in cheese samples during the winter months is very high, and that may be due to several factors, including the use of raw milk that contains large numbers of bacteria, and this leads to an increase number of bacteria in the resulting cheese and cream (10). Or, perhaps the reason is that it didn’t undergo enough thermal treatments to kill polluted microorganisms or not exposed to any heat treatments in most home-made cheese, small private fields or laboratories, in addition to marketing products in conditions that are free from health conditions and exposing cheese and cream in the air, which leads an increase in the content of microbes (1).

TABLE 2: Microorganisms were detected in the test sample

<table>
<thead>
<tr>
<th>Organism</th>
<th>No. of samples collected</th>
<th>No. of positive samples</th>
<th>% of positive samples</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>soft cheese</td>
<td>local cream</td>
<td>soft cheese</td>
</tr>
<tr>
<td>S. aureus</td>
<td>50</td>
<td>50</td>
<td>13</td>
</tr>
<tr>
<td>E.coli</td>
<td>50</td>
<td>50</td>
<td>10</td>
</tr>
<tr>
<td>Bacillus sp.</td>
<td>50</td>
<td>50</td>
<td>8</td>
</tr>
<tr>
<td>P.aeruginosa</td>
<td>50</td>
<td>50</td>
<td>5</td>
</tr>
<tr>
<td>K.pneumonia</td>
<td>6</td>
<td></td>
<td>5</td>
</tr>
<tr>
<td>Salmonella sp.</td>
<td>50</td>
<td>50</td>
<td>5</td>
</tr>
<tr>
<td>Yeast</td>
<td>50</td>
<td>50</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>50</td>
<td>4</td>
</tr>
</tbody>
</table>
When comparing these results with other studies, we find that they were similar with those identified by Alkhuzaai, 2006 (2), which documented that the total number of bacteria in soft cheese and local cream during the winter 6.3 x 10^6, 8.7 x 10^8. It is also noted from Table (2) that the emergence of strains of E.coli bacteria with average %20 in soft cheese and %24 in local cream samples, the presence of these bacteria in milk product samples indicates that these products are not clean, due to insufficient milk pasteurization or contamination of milk after pasteurization, especially fecal contamination and the presence of pathogenic bacteria.

It is clear from the same table the high rate of golden Staphylococcus bacteria 26% in soft cheese and 30% in local cream, and this percentage is located within the necessary limits for food poisoning events. This finding was similar to those reported by Singh and Prakash (21).

These rates may be due to the use of raw milk containing large numbers of golden clusters without suitable thermal treatment when making cheese and cream, as milk is taken from the cow or Buffalo infected with udder infection is an important source of contamination by golden clusters (4, 18).

The incidence of the species of E. coli itself in milk and milk products, as a possible cause of food born disease, is not significant if E. coli is normally a ubiquitous organism (13), the pathogenic strains if present could be harmful to consumers. The risk is magnified when the same samples of soft cheese and local cream are contaminated with S.aureus because this strain releases a toxic chemical enterotoxin. As little as 1.0 μg of the toxin in contaminated food produces symptoms of illness. This level of the toxin has been found at 105 cells /g of food (5).

Through Table (2) we notice the contamination of the soft cheese and local cream samples with Salmonella, as the cheese contamination rate reached 10%, while the cream pollution rate was 6% and explains the presence Salmonella sp. based on the use of raw milk is contaminated with large numbers of Salmonella that have been transmitted often comes from an infected cow or a carrier of the bacteria or from workers in the production of milk, moreover, lack of thermal treatments ensures that these bacteria will arrive in the milk, This results is similar with previous studies noted by Ekici K. et al. (8).

The other bacterial species identified from this particular study area included Bacillus sp. (16%), Pseudomonas aeruginosa and Klebsiella pneumonia (10%) for each genus in cheese samples, while Bacillus sp. (20%), Pseudomonas aeruginosa and Klebsiella pneumonia (6%) for each genus in local cream samples table 2. Most of the bacteria identified here are indicators of poor sanitary and hygienic condition of the farms and transportation system which is similar with previous report by Garedew et al. (12).
As for molds, they were averaged in cheese and cream 8%. Pollution with molds may be due to its ability to grow with soft cheese due to its high acidity and dairy contamination, including cheese or cream, may occur after the pasteurization process, because conducting this process is considered as the only determinant of the existence of this type of microorganism, or the reason may be due to the poor quality of the milk used (11&19).

Traditional practices are likely to contribute to the contamination of the milk and proliferation of the micro-organisms. The implication is that there is high risk of acquiring foodborne diseases since 36% of the residents have the habit of consuming raw or unpasteurized milk. It is therefore essential to implement control measures at each causes mentioned in our investigation.

CONCLUSIONS
Milk is an excellent nutrient medium for the growth of pathogenic bacteria that contaminate milk products from the various sources. The result of bacteriological assessment of milk products contaminants in this study showed that diverse environmental bacteria could contaminate milk and its products. Mastitis, unclean dairy houses, improper milking procedure and udder preparation, negligence on post-milking teat dipping and lack of herd health management all of these cause lead to the contamination of the milk and it is products.

We conclude from this study the poor quality of soft cheese and cream in the local markets of Al-Muthanna city given microbiological indicators exceed the internationally accepted limits, which indicates the inefficiency of transactions the heat used in the manufacture of these cheeses and cream, so we recommend that educating the dairy farm owners about microbial safety procedures, and must pay more attention and encourage responsible for the inclusion of soft cheese and local cream under health control conditions by setting specific standards for their production.

CONFLICT OF INTEREST

REFERENCES
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15. Karim S. Kh (2001): Isolate and diagnose some of the bacteria responsible for flavoring cheese