Get prevalence of Enterobacteriaceae in raw milk and some dairy products

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Abstract

Objective: This study was performed to detect and identify members of Enterobacteriaceae in raw milk, yoghurt and ice cream to assess their hygienic quality.

Methods: A total number of 200 samples distributed as follows: 100 raw milk, 25 small and 25 large scale plain yoghurt, and 25 small and 25 large scale ice cream samples collected from different localities at Kafr El-Sheikh Governorate and examined for Enterobacteriaceae count and identification of members.

Results: Enterobacteriaceae were detected in 84% of examined raw milk samples (with mean count of 1.02x10^6 + 1.98x10^5 cfu/ml), 40% and 32% of examined small and large scale plain yoghurt samples (with mean value of 6.95x10^3 + 4x10^3 and 1.57x10^3 + 8.95x10^2 cfu/g), while detected in 64% and 20% of examined small and large scale ice cream samples (with mean count of 1.02x10^4 + 4.13x10^3 and 1.4x10^3 + 8.26x10^2 cfu/g). The most frequent members isolated were Hafnia alvei (30.95%), E. coli (44.44%), and Serratia marcescens (57.14%) from positive raw milk, yoghurt, and ice cream samples, respectively. Other members of Enterobacteriaceae were isolated from positive samples at different percentages.

Conclusion: Presence of members of Enterobacteriaceae is a reliable index of fecal contamination and the possibility of the presence of associated enteric pathogenic organisms. So, it is recommended to specify Enterobacteriaceae as a sanitary index of milk and dairy products in the Egyptian food acts and regulations.

Keywords: Enterobacteriaceae, raw milk, yoghurt, ice cream

1. Introduction

Enterobacteriaceae is a large, heterogeneous group of Gram-negative rods, which naturally inhabit the intestinal tracts of both humans and animals. They include many genera (such as Escherichia coli, Shigella, Salmonella, Enterobacter, Klebsiella, Serratia, and Proteus). These genera are widespread in the environment and can contaminate milk through feces; bedding, improperly cleaned teats, milk handling, and equipment contaminated with soil or polluted water (Cohen et al., 2017). The presence of Enterobacteriaceae in dairy products induces undesirable changes that render the product of inferior quality, unmarketable, and unfit for human consumption. Moreover, their presence is frequently considered as a reliable index of fecal contamination. Therefore, the presence of Enterobacteriaceae and Coliform are routinely assessed to determine the hygienic quality of foods, particularly dairy products (Martin et al., 2010).

Food-borne microorganisms are major pathogens affecting food safety and cause human illness worldwide as a result of the consumption of foodstuffs, mainly animal products contaminated with vegetative pathogens or their toxins (Abebe et al., 2020). The outbreak of foodborne illnesses following consumption of raw milk and dairy products made from raw milk may be caused by Shiga toxin-producing Escherichia coli (STEC), Salmonella spp., and Yersinia enterocolitica (Proctor and Davis, 2000; Mazurek et al., 2004; Jayarao et al., 2006 and Cancino-Padilla et al. 2017). The primary condition associated with cases of foodborne illness caused by STEC and Salmonella spp. is gastroenteritis which is usually self-limiting, while immunocompromised individuals are at a higher risk of serious illness. The Yersinia enterocolitica disease is typically characterized by gastroenteritis and enterocolitis. However, debilitating post-infection immunological sequel, including Guillain-Barré syndrome and reactive arthritis are known to develop in some individuals following an episode of foodborne illness with these pathogens (Altekruse et al., 1999 and Oliver et al., 2005).

Yoghurt is the best known and popular cultured milk product concerning nutrition as it contains a high level of protein, calcium, phosphorus, and vitamin B12 and D3 (Piaia, 2001). Yoghurt helps to maintain food and water intake, which are often reduced in older people, moreover, it is a part of the diet designed to promote successful growth.

Ice cream is a highly delicious and nutritionally rich frozen milk product
that is widely consumed in Egypt. Microorganisms may gain entrance to such products during processing, handling and distribution also the ingredients used in the manufacture contribute to contamination of the product and consequently lead to public health hazards as gastroenteritis in humans (Yadav et.al. 1993 and Holban and Grumezescu, 2018).

Although pasteurization can destroy most of the specific pathogens that pose risk to public health, a potential microbial hazard may still be introduced after pasteurization through adding contaminated ingredients and improper handling process (Marshall, 1998 and Oliver, 2005). Raw milk that consumed raw or used in the manufacturing of yoghurt and ice cream may be produced under poor hygienic status using primitive procedures. Under such circumstances, the finished products would express to harbor a great number and different types of contaminants, particularly those belonging to enteric microorganisms. Therefore, this work was planned to detect and identify members of Enterobacteriaceae in raw milk, yoghurt, and ice cream to assess their quality and sanitation.

2. Materials and methods

This study was conducted after under the ethical approval from the Experimental Animals Care Committee in compliance with guidelines of the University of Kafrelsheikh.

2.1. Collection of samples

A total of 200 random samples of raw milk and some dairy products were collected from small dairies, groceries, and supermarkets from different localities at Kafr El Sheikh Governorate, Egypt. Of them, 100 raw milk, 50 plain yoghurt (25 small scale and 25 large scale), and 50 ice cream (25 small scale and 25 large scale) samples were obtained in their containers as sold to the consumer or in sterile and precipitated bile were counted and recorded as total Enterobacteriaceae count. Five colonies were picked up and isolated colonies were purified on plate count agar at 37°C/24 h and pathogenic organisms despite interference with acid development and the possible presence of enteric pathogens. Therefore, this work was planned to detect and identify members of Enterobacteriaceae in yoghurt and ice cream samples (>10 cfu/g) (Table 3) exceed the permissible limit of Egyptian Standards, 2005.

It is evident that the most frequent members isolated from the examined raw milk samples were Hafnia alive (30.95%), Serratia liquefaciens (25.0%), and Klebsiella pneumonia (15.48%). Klebsiella pneumoniae found in soil, water, sewage and constitute a part of the flora of the mouth and intestinal tract of human and animals. It responsible for pneumonia and upper respiratory tract infection and may be responsible for meningitis, pyaemia, and cystitis (Martin and Bachman, 2018). The prevalence of Klebsiella oxytoca, Enterobacter aerogenes and Proteus vulgaris was 13.1% (of the positive samples). Both Serratia marcescens and Proteus rettgeri were isolated at percentage of 11.9%, while, E. coli, Providencia rettgeri and Citrobacter diversus could be identified in 9.52, 9.52, and 3.57% of the positive samples, respectively.

Enterobacteriaceae members which isolated at low percentage were Proteus morganii, Providencia alcalifaciens, Citrobacter freundii, Shigella flexneri and Shigella sonnei each at 2.38% of the positive samples, while Enterobacter cloacae and Yersinia enterocolitica isolated only from 1.19% of the positive samples. Most of these groups were isolated by many investigators (Saleh, 2000; Nyein et. al. 2002 and Jayarao et. al. 2006).

3. Results and discussion

Enterobacteriaceae were detected in 84% (84%) of the examined raw milk samples with a mean count of 1.02 x 10^6 ± 1.98 x 10^5 cfu/ml. The highest frequency distribution (48.81%) of the positive samples lies within the range of 10^5: <10^6 cfu/ml followed by 22.62% in the range of 10^6: <10^7 cfu/ml (Tables 1 and 2).

Enterobacteriaceae members were detected in 40% and 32% of the examined small scale and large scale plain yoghurt samples with a mean value of 6.95 x 10^3 ± 4 x 10^3 and 1.57x10^4 ± 8.95 x10^3 cfu/g (Table 1). The highest frequency distribution of positive small scale plain yoghurt samples (70%) lies within the range of 1 x 10^3:<5 x 10^4 cfu/g while that of positive large scale plain yoghurt samples (62.5%) lies within the range of >10^5:<10^6cfu/g (Table 3).

On the other hand Enterobacteriaceae members were detected in 64% and 20% of the examined small scale and large scale ice cream samples with a mean count of 1.02 x 10^5 ±4.13 x 10^3 and 1.4 x 10^5 ±8.26 x 10^3 cfu/g (Table 1). The highest frequency distribution of both small scale ice cream samples (43.75%) and large scale ice cream samples (60%) lie within the range of >10^5:<10^6 cfu/g (Table 3). According to Egyptian Standards for yoghurt and ice cream, Enterobacteriaceae count must not exceed 10 cell/g (E.S.2005, a and b), consequently all positive yoghurt and ice cream samples (>10 cfu/g) (Table 3) exceed the permissible limit of Egyptian Standards, 2005.

The presence of a large number of Enterobacteriaceae in raw milk indicate unsafe raw milk for human consumption (Nyein et al. 2002) unless it is pasteurized as Enterobacteriaceae in raw milk were completely removed by pasteurization and not appear during the manufacturing process of dairy product (Branciareti et. al. 2004) so milk is the most frequent member of the large bowel of man and
animal, thus its presence in food generally indicates direct or indirect fecal pollution, so that it has been implicated in cases of gastroenteritis, epidemic diarrhea in infants, sporadic cases of summer diarrhea in children and cases of food poisoning (Bennett et al., 2015). It is important to conclude that the detection of even low numbers of E. coli in foods reveals public health risk (I.C.M.S.F., 1978). According to Egyptian Standards, yogurt must be free from E. coli (E.S. 2005; a), thus confirm public health risk for yoghurt consumers.

Serratia liquefaciens could be isolated from 27.78% of the positive samples examined while Shigella sonnei, Shigella flexneri, Klebsiella pneumoniae, and Hafnia alvei isolated at 22.22%. On the other hand, Serratia marcescens, Enterobacter aerogenes, Providencia stuartii, Edwardsiella hoshinae, and Proteus morganii were detected in 11.11%. Citrobacter diversus, Proteus rettgerii, and Enterobacter cloacae were detected in 5.56% of the positive samples. No salmonellae could be detected in any of the examined yoghurt samples. Similar results were recorded by many authors (El-Kasas, 2004; Braun and Preuss, 2007 and Rotar et al., 2007).

Identification of Enterobacteriaceae isolated from ice cream samples as recorded in Table (6). Serratia marcescens had the highest isolation percentage (57.14% out of 21 positive samples). It was reported that Serratia organisms have been implicated in human infection, including pulmonary and urinary tract infections as well as septicemia (Mahlen, 2011). E. coli could be isolated at 33.33% which is nearly similar to results recorded by (Yaman et al., 2006). Both Klebsiella pneumoniae and Hafnia alvei were isolated from 23.81% while both Citrobacter diversus and Enterobacter aerogenes isolated with a percentage of 19.05%. On the other hand, Klebsiella ozaenae, Citrobacter freundii, Proteus rettgerii, and Shigella flexneri were detected in 9.52% of positive samples and Serratia odorifera, Enterobacter cloacae, Shigella sonnei, and Proteus vulgaris were isolated only from one sample at a percentage of 4.76 of the positive samples. This study showed that Salmonella spp. could not be detected in any of the examined ice cream samples as reported by many previous authors (Fadel and Ismail, 2009 and El-Bana, 2011).

The high Enterobacteriaceae count in the examined ice cream samples obtained in this investigation reflects unhygienic practices during manufacture and distribution. This may be due to poor ingredients, carelessness of the employer, who may lack the necessary knowledge of personal hygiene, product handling, and sanitation routines. So, this indicates a need for more concern on the part of the dairy industry to prevent contamination of ice cream. Ensure ingredients of good quality and especially those added after heat treatment should be free from pathogens and conform to microbiological criteria. Sufficient heat treatment to ice cream mix, prevention of post heat treatment contamination, and special attention should be given to utensils and equipment which come in contact with ice cream also the packaging used.

Enterobacteriaceae are sometimes contaminants of pasteurized dairy products. Their presence is indicative of unsanitary methods of manufacture, inadequate pasteurization, or post-pasteurization contamination. This indicates a need for more concern on the part of the dairy industry to prevent contamination with Enterobacteriaceae and thus prevent additional outbreaks of food-borne illness caused by their members.

Table 1. Statistical analytical results of Enterobacteriaceae count in the examined samples

| Type of samples | No. of examined samples | Positive samples | Enterobacteriaceae count (cfu/ml or g) |
|-----------------|-------------------------|-----------------|--------------------------------------|
|                 |                         | No. | %     | Minimum | Mean ± SE          |
| Raw milk        | 100                     | 84  | 84    | 3 x 10^2 | 1.02 x 10^6 ± 1.98 x 10^5 |
| Yoghurt         |                         |     |       |         |                   |
| -Small scale    | 25                      | 10  | 40    | 9 x 10^2 | 6.95 x 10^3 ± 4 x 10^3 |
| -large scale    | 25                      | 8   | 32    | 5 x 10  | 1.57 x 10^3 ± 8.95 x 10^2 * |
| Ice cream       |                         |     |       |         |                   |
| -Small scale    | 25                      | 16  | 64    | 5 x 10  | 1.02 x 10^4 ± 4.13 x 10^3 |
| -large scale    | 25                      | 5   | 20    | 3 x 10^2 | 1.4 x 10^3 ± 8.26 x 10^2 |

cfu = colony forming unit, SE= Standard error of mean
* Small and large scale (yoghurt and ice cream) samples differed significantly at P < 0.01

Table 2. Frequency distribution of positive raw milk samples based on their Enterobacteriaceae count

| Interval (cfu/ml) | No. of samples | %     |
|------------------|----------------|-------|
| >10^3 : < 10^4   | 7              | 8.33  |
| 10^4 : < 10^5    | 16             | 19.05 |
| 10^5 : < 10^6    | 41             | 48.81 |
| 10^6 : < 10^7    | 19             | 22.62 |
| ≥ 10^7           | 1              | 1.19  |
| Total            | 84             | 100   |
Table 3. Frequency distribution of positive yoghurt and ice cream samples based on their Enterobacteriaceae count

| Interval (cfu/g) | Yoghurt samples | Ice cream samples |
|----------------|----------------|------------------|
|                | No. | %   | No. | %   | No. | %   | No. | %   |
| >10^2: < 10^3  | 1   | 5.6 | 7   | 25.0 | 3   | 27.75 | 3   | 27.75 |
| 10^3: < 5 x 10^3 | 7   | 25.0 | 2   | 10.0 | 5   | 27.75 | 5   | 27.75 |
| 5 x 10^3: < 10^4 | 1   | 3.8 | 1   | 5.3 | 1   | 3.82 | 0   | 0   |
| 10^4: < 5 x 10^4 | 1   | 3.8 | 0   | 0   | 0   | 0   | 0   | 0   |
| Total          | 10  | 100 | 8   | 100 | 16  | 100 | 5   | 100 |

Table 4. Incidence of Enterobacteriaceae members isolated from the examined raw milk samples

| Isolates             | Number | % * | Positive samples |
|----------------------|--------|-----|-----------------|
| Hafnia alve           | 54     | 17.65 | 26       | 30.95 |
| Serratia liquefaciens | 34     | 11.11 | 21       | 25.00 |
| Klebsiella pneumoniae | 18     | 5.88  | 13       | 15.48 |
| Klebsiella oxytoca    | 30     | 9.80  | 11       | 13.10 |
| Enterobacter aerogenes| 20     | 6.54  | 11       | 13.10 |
| Proteus vulgaris      | 12     | 3.92  | 11       | 13.10 |
| Serratia marcescens   | 44     | 14.38 | 10       | 11.90 |
| Proteus rettgerii     | 28     | 9.15  | 10       | 11.90 |
| Escherichia coli      | 20     | 6.54  | 8        | 9.52  |
| Providencia rettgeri  | 10     | 3.27  | 8        | 9.52  |
| Citrobacter diversus  | 4      | 1.31  | 3        | 3.57  |
| Proteus morganii      | 3      | 0.98  | 2        | 2.38  |
| Providencia alcalifaciens | 4     | 1.31  | 2        | 2.38  |
| Citrobacter freundii  | 8      | 2.61  | 2        | 2.38  |
| Shigella flexneri     | 6      | 1.96  | 2        | 2.38  |
| Shigella sonnei       | 6      | 1.96  | 2        | 2.38  |
| Enterobacter cloacae  | 4      | 1.31  | 1        | 1.19  |
| Yersinia enterocolitica | 1    | 0.33  | 1        | 1.19  |
* Percent calculated to total No. of isolates (306)  ** Percent calculated to number of positive samples (84)

Table 5. Incidence of Enterobacteriaceae members isolated from the examined yoghurt samples

| Isolates             | Number | % * | Positive sample |
|----------------------|--------|-----|-----------------|
| E. coli              | 42     | 26.58 | 8          | 44.44 |
| Serratia liquefaciens | 20     | 12.66 | 5          | 27.78 |
| Shigella flexneri    | 14     | 8.86  | 4          | 22.22 |
| Shigella sonnei      | 22     | 13.92 | 4          | 22.22 |
| Klebsiella pneumoniae | 12    | 7.59  | 4          | 22.22 |
| Hafnia alve           | 10     | 6.33  | 4          | 22.22 |
| Serratia marcescens   | 10     | 6.33  | 2          | 11.11 |
| Enterobacter aerogenes| 8      | 5.06  | 2          | 11.11 |
| Providencia staurtii  | 4      | 2.53  | 2          | 11.11 |
| Edwardsiella hoshinae| 2      | 1.27  | 2          | 11.11 |
| Proteus morganii      | 2      | 1.27  | 2          | 11.11 |
| Proteus rettgerii     | 2      | 1.27  | 1          | 5.56  |
| Enterobacter cloacae  | 8      | 5.06  | 1          | 5.56  |
| Citrobacter diversus  | 2      | 1.27  | 1          | 5.56  |
* Percent calculated to total No. of isolates (158)  ** Percent calculated to number of positive samples (18)

Table 6. Incidence of Enterobacteriaceae members isolated from the examined ice cream samples

| Isolates             | Number | % * | Positive samples |
|----------------------|--------|-----|-----------------|
| Serratia marcescens   | 24     | 21.82 | 12         | 57.14 |
| Escherichia coli      | 16     | 14.55 | 7          | 33.33 |
| Klebsiella pneumoniae | 14     | 12.73 | 5          | 23.81 |
| Hafnia alve           | 12     | 10.91 | 5          | 23.81 |
| Citrobacter diversus  | 8      | 7.27  | 4          | 19.05 |
| Enterobacter aerogenes| 8      | 7.27  | 4          | 19.05 |
| Klebsiella ozaena     | 4      | 3.64  | 2          | 9.52  |
| Citrobacter freundii  | 4      | 3.64  | 2          | 9.52  |
| Shigella flexneri     | 6      | 5.45  | 2          | 9.52  |
| Proteus rettgerii     | 6      | 5.45  | 2          | 9.52  |
| Serratia odorifera    | 2      | 1.82  | 1          | 4.76  |
| Enterobacter cloacae  | 2      | 1.82  | 1          | 4.76  |
| Shigella sonnei       | 2      | 1.82  | 1          | 4.76  |
| Proteus vulgaris      | 2      | 1.82  | 1          | 4.76  |
* Percent calculated to total No. of isolates (110)  ** Percent calculated to number of positive samples (21)
4. Conclusion
The results obtained allow concluding that the sanitary measures adopted during production, handling, and distribution of the examined milk and milk products (yoghurt and ice cream) are neglected in most cases as Enterobacteriaceae members existed in most samples examined. Also, the presence of these members of Enterobacteriaceae is a reliable index of fecal contamination and the possibility of the presence of associated enteric pathogenic organisms. So, Good hygiene, GMP, sanitation in operating procedures, and implementation of standardized HACCP and pasteurization procedures are effective methods for control and prevention. Therefore, it is recommended to specify the Enterobacteriaceae as a sanitary index of milk and dairy products in the Egyptian food acts and regulations.

References
Abebe E, Gugsa G and Ahmed M (2020): Review on Major Foodborne Zoonotic Bacterial Pathogens. J.Trop.Med. vol. 2020. https://doi.org/10.1155/2020/4674235.

American Public Health Association (A.P.H.A.) (1992): Standard methods for examination of dairy products. 16th Ed., New York. USA.

American Public Health Association (A.P.H.A.) (1992): Standard methods for the examination of dairy products. 17th Ed., New York. USA.

Alkekruse SF, Stern NJ, Fields PI and Swerdlow DL (1999): Campylobacter jejuni-an emerging food born pathogen. Emerg. Infect. Dis.,5: 28-35.

Bennett JE, Dolin R and Blaser MJ (2015): Mandell, Douglas, and Bennett’s principals and practice of infectious diseases,8 th Ed.

Branciari R, Goga BTC, Rea S and Avellini P (2004): Evaluation of E.S. (2005, b): Egyptian Organization for Standardization and Quality. El-Kasas, Walaa MA (2004): Microbiological studies on fermented milk and milk products in Yangon during 2000, Myanmar. Health. Sci. Res. J., 14 (1/3): 35-41.

Oliver SP, Jayarao BM and Almeida RA (2005): Food borne pathogens in milk and the dairy farm environment: Food Safety and Public Health Implications. Food Borne Pathog. Dis. 2005; 2: 115-129.

Sobeih et al., 2020, KVMJ, 18 (2): 9-13, DOI: 10.21608/kvmj.2020.39992.1009

Braun G and Preuss SE (2007): Microbial quality of water buffalo milk and milk products in Germany. Milchwissenschaft; 62 (3): 276-278.

Cancino-Padilla N, Fellenberg MA, Franco W, Ibanez RA and Vargas-Bello-Perez E (2017): Foodborne bacteria in dairy products: Detection by molecular techniques. Cien.Inv.Agr., 215-229.

Cohen I, Powderly WG and Opal SM (2017): Infectious diseases, 4 th Ed. Elsevier Ltd.

El-Bana, Amal FA (2011): Bacteriological Studies on Ice-Cream in Kafr El-Sheikh Governorate. M. V. Sc. Thesis, Fac. Vet. Med. Kafrelsheikh Univ.

El-Kasas, Walaa MA (2004): Microbiological studies on fermented milks in Kafr El-Sheikh Governorate. M. V. Sc. Thesis, Fac. Vet. Med. Tanta Univ.

E.S. (2005, a): Egyptian Organization for Standardization and Quality. Yoghurt, ES: 1000/2005.

E.S. (2005, b): Egyptian Organization for Standardization and Quality. Milk and water ice (Ice cream) Part: 1, ES: 1185-1/2005.

Fadel HM and Ismail, J (2009): Prevalence and significance of Staphylococcus and Enterobacteriaceae species in selected dairy products and handlers. Inter. J. Dairy Sci.; 4(3):100-108.

Harrigan WF (1998): Laboratory methods in food microbiology, 3rd Ed., Academic Press, San Diego, California 92101-4495, USA.

International Commission for Microbiological Specification for Food (I.C.M.S.F) (1978): Microbial ecology of foods. Vol. II. Academic Press, N.Y.

Jayarao BM, Donaldson SC, Straley BA, Sawant AA, Hegade NV and Brown JL (2006): A survey of food borne pathogens in bulk tank milk and raw milk consumption among farm families in Pennsylvania. J. Dairy Sci., 89: 2451-2458.

Krieg NR and Holt JG (1984): Bergey’s manual of systematic bacteriology. Vol. 1. Williams and Wilkins. Baltimore, London.

Mahlen SD (2011): Serratia Infections from Military Experiments to current practice. Clin. Microbiol. Rev., 24(4), 755-791.

Marshall RT (1998): Ice cream and frozen yoghurt. Applied dairy microbiology. Edited by Marth, EW. and Streele, JL. Marcel Dekker. New York.

Martin MC, Martinez N, del Rio B, Ladero V, Fernandez M and Alvarez MA (2010): A novel real-time polymerase chain reaction-based method for the detection and quantification of lactose-fermenting Enterobacteriaceae in the dairy and other food industries. J. Dairy Sci., 93(3): 860-867.

Martin RM and Bachman MA (2018): Colonization, infection, and the Accessory Genome of Klebsiella pneumoniae. J. Front. cell. Infig. Microbial., 8(4).

Mazurek J, Salehi E, Propes D, Holt J, Bannerman T, Nicholson LM, Bundesen M, Duffy R and Moolenaar RL (2004): A multistate outbreak of Salmonella enterica serotype typhimurium infection linked to raw milk consumption-Ohio, 2003. J. Food Prot., 67: 21-65.

Nyein; Mar-Mar, Khine TT and Thin KK (2002): Bacteriological aspects of milk and milk products in Yangon during 2000. Myanmar. Health. Sci. Res. J., 14 (1/3): 35-41.

Oliver SP, Jayarao BM and Almeida RA (2005): Food borne pathogens in milk and the dairy farm environment: Food Safety and Public Health Implications. Food Borne Pathog. Dis. 2005; 2: 115-129.

Piaia M (2001): Fermented milk and successful aging. Danone World News Letter No. 22: 1-14.

Proctor ME and Davis JP (2000): Escherichia coli O157: H7 Infections in Wisconsin 1992-1999 Wis. Med. J., 99: 32.

Reed BA and Grivetti LE (2000): Controlling on farm inventories of bulk tank raw milk an opportunity to protect Public Health. J. Dairy Sci., 83: 2988 (Abstract).

Rotar MA, Semeniuc C, Apostu S, Suharoschi R, Muresan C, Modooran C, Laslo C, Gus C and Gulea M (2007): Researches concerning microbiological evolution of lactic acid bacteria to yoghurt storage during shelf-life, J. Agric., USAMV Cluj-Napoca, Cluj Napoca, Romania, J. Agroalimentary Processes and Technol., 13(1): 135-138.

Saleh, Ormaye A (2000): Bacteriological studies on some low-temperature gram-negative bacteria in milk and some dairy products. Egypt. J. Agric. Res., 78 (1, Special Issue): 273-277.

Yadav JS, Grover S and Batish VK (1993): Comprehensive dairy microbiology. Published by Metropolitan Book Co. Pvt Ltd. New Delhi, 1 st Ed., Ch. ON Microbiology of Ice Cream, 259-283.

Yaman H, Elmal M, Ulukanl Z, Tuzcu M, Genctav K (2006): Microbial quality of ice cream sold openly by retail outlets in Turkey. Rev. de Med. Vet., 157(10): 457-462.