

## MICROBIOLOGICAL Quality OF MILK AND ICE CREAM SOLD IN KAFR EL-SHEIKH AND EL- GHARBIA GOVERNORATES

By

Azza, M.K. Sobeih; I.I. Al-Hawary and I. Aman

Dept. of Food Hygiene, Fac. Vet. Med. Kafr El-Sheikh. Tanta Univ.

### SUMMARY

*One hundred and forty samples (50 raw milk, 45 UHT milk and 45 packed ice cream) collected randomly from different markets at kafr El-Sheikh and El-Gharbia Governorates were examined microbiologically. In addition, UHT milk samples were examined for visual, organoleptic and for K.Q. tests. All raw milk samples (100%) contained aerobic bacteria with a mean count of  $2.9 \times 10^8$  cfu/ml while 96%, 88%, 94% and 88% of the examined samples were contaminated with coliform, thermotolerant, psychrotrophic and E.coli bacteria with mean values of  $5.6 \times 10^7$  MPN/ml,  $1.9 \times 10^2$ ,  $7.4 \times 10^7$  and  $5.4 \times 10^6$  cfu/ml respectively. While all UHT milk samples showed no swelling or organoleptic changes and satisfied the acidity and APT. Microbiologically, they were contaminated with aerobic bacteria (100%), anaerobic spore formers (11%) and B.cereus (17.8% of samples) with mean counts of  $3.1 \times 10^4$ ,  $1.5 \times 10^2$  and  $2.3 \times 10^2$  cfu/ml respectively. Examination of ice cream samples revealed that 2 (4.4%) of samples were contaminated with salmonella and all samples (100%) were contaminated with aerobic bacteria, staph. aureus and psychrotrophic bacteria with average counts of  $1.4 \times 10^7$ ,  $6.8 \times 10^5$  and  $8.3 \times 10^4$  cfu/g respectively.*

## **INTRODUCTION**

Milk and ice cream are widely consumed by infants and children worldwide. They are liable to contamination by both pathogenic and spoilage microorganisms from different sources during production, handling and distribution (Ahmed and Sallam, 1991).

Raw milk in urban places sold raw and sometimes may be consumed raw or processed into raw milk products. Hence, it is responsible for serious diseases especially if produced from diseased animals.

In Egypt, ultra heat treated (UHT) milk has gained increase acceptance during the last few years to safeguard the milk consumers as well as to overcome the lack of cooling facilities needed by pasteurized milk at retail outlets. In spite of its subjection to high temperature during processing several investigators (Langefeld and Bolle 1979, Farahnik 1982 and El-Talawy 1998) could isolate aerobic and anaerobic spore formers organisms that render the product unfit for consumption and reflect something error in measures adopted during processing or post-processing. The majority of post-processing contamination is related to packaging problem or due to failure of equipment sterilization downstream processing.

Moreover, in summer a wide range of people of all ages consume ice cream. The product may be subjected to contamination either from ingredients used or during processing. Numerous epidemics and food poisoning outbreaks have been attributed to the consumption of contaminated ice cream (Bryan, 1981 and Galbraith et al., 1982). Therefore, this work was planned to study the microbial criteria of milk and ice cream and the effect of contaminating microorganisms on their quality.

## **MATERIAL AND METHODS**

### **Samples: -**

One hundred and forty samples (50 raw milk, 45 UHT milk and 45 packed ice cream) were purchased in retail packages from different markets at kafr EL-

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Sheikh and El-Gharbia Governorates, Samples were transferred to the laboratory and examined on the same day.

**Preparation and examination of samples: -**

Raw milk samples: -

Collected samples were prepared and enumerated for aerobic bacteria, coliform bacteria, thermophilic bacteria and psychrotrophic bacteria according to APHA (1992). E.coli was counted, isolated and identified according to APHA (1992).

UHT milk samples: -

To detect unsterile package, collected samples were pre-incubated at 30°C for 5 days (IDF, 1972). Then each package was examined visually for swelling, organoleptic properties, titratable acidity, ethanol (68% v/v) stability test and microbiologically for aerobic bacteria and anaerobic spore former bacteria according to APHA, (1992). B.cereus organism was counted, isolated and identified according to Adams and Moss(1995).

Ice cream samples: -

Collected samples were melted at 15°C and counted for aerobic and psychrotrophic bacteria. Staphylococcus aureus was counted, isolated and identified according to APHA (1992). Moreover, isolation of salmonella was carried out on S.S agar media after enrichment on Selenite-F broth and presumptive identification of salmonella from other enterobacteriaceae was carried out by using biochemical tests described by Cowan and Steel (1974).

## **RESULTS and DISCUSSION**

The microbiological criteria of raw milk samples are presented in table (1). All samples (100%) analyzed contained aerobic bacteria in counts ranged from  $4 \times 10^4$  to  $2 \times 10^9$  with a mean value of  $2.9 \times 10^8$  cfu/ml. Seventy eight percent of the positive samples exceeding the allowable limit of  $< 10^5$  cfu/ml for ABC (table 5) which indicates serious faults in production and handling hygiene or due to lack of cooling facilities during transportation as total aerobic counts are usually used to assess the overall sanitation and storage conditions of raw milk. Thus, all dairy

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regulations include the use of total aerobic counts to grade milk for the producer quality payment scheme.

The presence of large numbers of coliform bacteria in milk also generally provides an index of the hygienic standards used in the production of milk, as unclean udders and teats can contribute coliforms from a variety of sources such as manure, soil, feed and water (Richter et al., 1992). Ninety six percent of the examined raw milk samples contaminated with coliforms in counts ranged from  $4 \times 10^4$  to  $2 \times 10^8$  with a mean count of  $5.6 \times 10^7$  MPN/ml (table1). Similar counts were recorded by El-Shinawy et al. (1995a) and Deeb (1996). Moreover, 95.8% of coliform positive samples containing counts more than the allowable limit of  $<10^2$  (table 5). Such milk with large coliform numbers may carry entero-pathogenic bacteria such as salmonella species that may result in human infection and disease (Fontaine et al., 1980).

It is also clear from table (1) that thermotolerant organisms were detected in 88% of samples with a mean count of  $1.9 \times 10^2$ , similar results were recorded by Kikuchi et al. (1996), while higher results were reported by Abd El-Ghani (1993). Only 6.8% of positive samples exceed the proposed upper limit of  $<10^3$  cfu/ml (table 5). Psychrotrophic organisms were detected in 94% of the examined samples with a mean value of  $7.4 \times 10^7$  cfu/ml, which is nearly similar to results recorded by Girgis et al. (1996). All the psychrotrophic positive samples exceed the allowable limit of  $<5 \times 10^2$  cfu/ml proposed by Luck (1972). Psychrotrophs are the main spoilage bacteria of milk kept at refrigeration temperature. They can grow, multiply and produce enzymes that degrade milk protein and fat resulting in technological problems during processing and reduce quality of pasteurized products. Moreover, pathogens as *L.monocytogenes*, *Aeromonas* and *St.zooepidemicus* are psychrotrophic organisms.

*E.coli* was isolated from 88% of raw milk samples with an average count of  $5.4 \times 10^6$  (table1). The high *E.coli* count indicates unhygienic measures during production and distribution. In addition, its presence in milk imply risk for consumer

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due to possible presence of enteric pathogens, rather than the public health importance of E.coli as Enteropathogenic stereotypes have been implicated in cases of gastroenteritis, epidemic diarrhea in infants, Traveler's diarrhea and food poisoning.

Results recorded in table (2) reveal that all UHT milk samples after pre-incubation at 30°C for 5 days were with normal visual and organoleptic properties (sweet, no swelling) and stable with ethanol 68% as well as the titratable acidity was within the normal limit.

Aerobic bacteria were detected in all (100%) of the examined UHT milk samples with counts ranged from  $1 \times 10^2$  to  $9 \times 10^4$  cfu/ml (table 3). Nearly similar findings were reported by Zahran (1999). All samples exceed the Egyptian allowable limit of  $< 10$  cfu/ml (table5). Failure of equipment sterilization downstream the thermal process or errors in packaging or storage are the main points of contamination. As the product was kept at refrigeration temperature these bacteria may be responsible for rapid deterioration due to their proteolytic activity (Shahin and Fahmy, 1988).

Anaerobic bacteria are of great importance as their presence in large numbers constitute a public health risk and responsible for several outbreaks. They are detected in 5(11%) of UHT milk samples examined with an average count of  $1.5 \times 10^2$  (table3). Hence, their presence in UHT milk may indicates failure in heat treatment or unsanitary practices after processing.

B.cereus bacteria could be detected in 8 (17.8%) of the examined UHT milk samples with an average count of  $2.3 \times 10^2$  cfu/ml (table3). Nearly similar result was recorded by El-Shinawy et. al., (1995 b), while higher percentage was recorded by Zahran (1999). It is the most important bacillus species of aerobic spore formers that has been reported to be a cause of many defects in heat-treated milks as bitty cream and sweet curdling. Moreover, it plays a role as a food borne pathogen responsible for food poisoning since 1950s (Geopfert et al., 1992).

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Ice cream samples showed ABC ranged from  $1 \times 10^4$  to  $9 \times 10^7$  cfu/g with an average of  $1.4 \times 10^7$  cfu/g (table 4). Nearly similar results were recorded by Rahul et al. (2000) who mentioned that these counts are higher than the safety limits prescribed by Indian standards institute, which indicates poor quality production.

Staphylococcus aureus organism was detected in 45(100%) of the examined ice cream samples with an average of  $6.8 \times 10^5$  cfu/g (table 4), the result which is higher than that reported by Patwari and Chavan (1995) and Abo-Risha (1998). All positive samples exceed the limit of  $<10^2$  cfu/g proposed by ICMSF, 1974 for Staph. aureus (table 5). These results allow us to conclude that ice cream sold in both Governorates may represent a risk from food poisoning.

Psychrotrophic bacteria as shown in table (4) ranged from  $9 \times 10$  to  $9 \times 10^5$  with a mean value of  $8.3 \times 10^4$ . This indicates unhygienic measures during processing, packaging and storage in addition to liability of these samples for spoilage during storage.

Salmonella was detected in 2(4.4%) of the examined ice cream samples that represent a public health hazard arising from risk of food poisoning infection.

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**Table (1) Microbiological criteria of the examined raw milk samples (n=50)**

Criteria	+ve samples		Min.	Max.	Mean
	No.	%			
Aerobic bacterial count (ABC) cfu/ml	50	100	$4 \times 10^4$	$2 \times 10^9$	$2.9 \times 10^8$
Coliform count (MPN/ml)	48	96	$4 \times 10$	$2 \times 10^8$	$5.6 \times 10^7$
Thermotolerant count (cfu/ml)	44	88	$1 \times 10$	$2.3 \times 10^3$	$1.9 \times 10^2$
Psychrotrophic count (cfu/ml)	47	94	$2 \times 10^3$	$4 \times 10^8$	$7.4 \times 10^7$
E-coli (cfu/ml)	44	88	$4 \times 10$	$1 \times 10^8$	$5.4 \times 10^6$

**Table (2) visual, organoleptic and K.Q. tests of UHT milk samples after pre-incubation at 30°C for 5 days**

Tests	Comment
Visual and organoleptic	All samples examined were sweet, without off-flavor and showed no swelling.
Keeping quality (K.Q.) (1) Acidity using titratable method (2) Ethanol stability	-It ranged from 0.13 to 0.15 with an average of 0.14 lactic acid % - All samples were stable and showed no deterioration



**Table (3) Microbiological criteria of the examined UHT milk samples (n=45)**

Criteria	+ve samples No.	%	Min.	Max.	Mean
<b>Aerobic bacterial count (cfu/ml)</b>	45	100	$1 \times 10^2$	$9 \times 10^4$	$3.1 \times 10^4$
<b>Anaerobic spore count</b>	5	11	$3 \times 10$	$4.9 \times 10^2$	$1.5 \times 10^2$
<b>B.cereus count</b>	8	17.8	$1 \times 10$	$1.2 \times 10^3$	$2.3 \times 10^2$

**Table (4) Microbiological criteria of the examined ice cream samples (n=45)**

Criteria	+ve samples No.	%	Min.	Max.	Mean
<b>Aerobic bacterial count (cfu/g)</b>	45	100	$1 \times 10^4$	$9 \times 10^7$	$1.4 \times 10^7$
<b>Staph.aureus count* (cfu/g)</b>	45	100	$9 \times 10^3$	$2.8 \times 10^6$	$6.8 \times 10^5$
<b>Psychrotrophic count (cfu/g)</b>	45	100	$9 \times 10$	$9 \times 10^5$	$8.3 \times 10^4$

N.B Salmonella was detected in 2(4.4%) of the examined ice cream samples.

\*Staphylococcus aureus count was based on its character on Baird Parker agar.

Table (5) Number of samples exceeding the maximum allowable limits abroad

Type of sample	Criteria	No. of +ve samples	Allowable limit/ml or g	No. of samples exceeding limit*	References
Raw milk	Aerobic bacterial count (cfu/ml)	50	$<10^5$	39 (78%)	IDF (1996)
	Coliform (MPN/ml)	48	$<10^2$	46(95.8%)	Luck (1972)
	Thermoduric count (cfu/ml)	44	$<10^3$	3 (6.8%)	
	Psychrotrophic count (cfu/ml)	47	$<5 \times 10^2$	47 (100%)	
	E-coli (cfu/ml)	44	$<10$	44 (100%)	
UHT milk	Aerobic bacterial count (cfu/ml)	45	$<10$	45(100%)	ES (1990)
	Anaerobic spore former	5	$<1$	5 (100%)	IDF (1972)
	B.cereus	8	$<1$	8 (100%)	
Ice cream	Aerobic bacterial count (cfu/g)	45	$<10^4$	43(95.6%)	ICMSF (1974)
	Staph.aureus	45	$<10^2$	45(100%)	
	Psychrotrophic	45	$<5 \times 10^2$	34(75.6%)	
	Salmonella	2	Absent in 25 g	2 (100%)	

\*from +ve samples

## الملخص العربي

### الجودة الميكروبيولوجية للألبان والأيس كريم المباعة بمحافظة كفر الشيخ والغربية

عزة محمود كامل صبيح – ابراهيم ابراهيم الهوارى – ابراهيم محمد أمان

تم تجميع مائة وأربعون عينة ( ٥٠ عينة ألبان خام، ٤٥ عينة لبن معقم و ٤٥ عينة ايس كريم معلبة ) من اسواق المراكز المختلفة بمحافظتى كفر الشيخ والغربية لفحصها. وقد أظهرت النتائج احتواء جميع عينات الألبان الخام على البكتيريا الهوائية بمتوسط عددي ٩ و ٢ و ١٠ × <sup>٨</sup> بينما اوضحت النتائج تلوث ٩٦% ، ٨٨% ، ٩٤% و ٨٨% من العينات بالميكروبات العسوية القولونية، البكتيريا المقاومة للبسترة، البكتيريا المحبة للبرودة وميكروب الأشيريشا كولاى بمتوسط عددي ٦ و ١٠ × <sup>٥</sup> ، ٩ و ١٠ × <sup>٢</sup> ، ٧ و ١٠ × <sup>٧</sup> و ٤ و ١٠ × <sup>٦</sup> على التوالي. فى حين أوضحت النتائج احتواء جميع عينات الألبان المعقمة على البكتيريا الهوائية بمتوسط او ٣ و ١٠ × <sup>٤</sup> ووجد ان ١١% من العينات ملوثة بالبكتيريا الاهوائية المتجرثمة بمتوسط ٥ و ١٠ × <sup>١</sup> وميكروب الباسيلس سيرس تم عزله من ٨ و ١٧% من العينات بمتوسط ٣ و ١٠ × <sup>٢</sup>. تبين من فص عينات الأيس كريم تلوث جميع العينات بالبكتيريا الهوائية والميكروب العنقودي الذهبى والبكتيريا المحبة للبرودة بمتوسط عددي ٤ و ١٠ × <sup>٧</sup> ، ٨ و ١٠ × <sup>٦</sup> و ٣ و ١٠ × <sup>٤</sup> على التوالي. بينما تم عزل ميكروب السالمونيلا من عينتين من عينات الأيس كريم.