



Evaluation of the bacteriological quality of ice cream sold in Trinidad

Anil Pooran, Nadira Seepersadsingh, Karla Georges and Abiodun A. Adesiyun *

School of Veterinary Medicine, Faculty of Medical Sciences, Eric Williams Medical Sciences Complex, University of the West Indies, St. Augustine Campus, Republic of Trinidad and Tobago. *e-mail: abiodunadesiyun@sta.uwi.edu

Received 4 January 2012, accepted 20 April 2012.

Abstract

The bacteriological quality of ice cream sold to consumers in Trinidad was evaluated and factors that may contribute to its contamination were assessed. The cross-sectional study was conducted on ice cream sold by supermarkets and street vendors across Trinidad. The frequency of *Salmonella* spp., *Listeria* spp., *Escherichia coli* and staphylococci, as well as the total aerobic plate counts (TAPC) per g of ice cream were determined using standard methods. The frequency of isolation of these bacteria was compared for imported and locally produced ice cream, commercial and home-made, flavour, temperature and pH of the product. Of a total of 115 ice cream samples comprising 52 foreign commercial, 38 local commercial and 25 local home-made, all were negative for *Salmonella* and *Listeria* spp. while 1 (1.9%), 8 (21.1%) and 13 (52.0%), respectively, were positive for staphylococci; and 2 (3.8%), 10 (26.3%) and 22 (88.0%), respectively, were contaminated by *E. coli*. The differences were statistically significant ($P < 0.05$). Overall, the TAPC per g of ice cream was $1.5 \times 10^2 \pm 2.9 \times 10^2$ for foreign commercial, $7.4 \times 10^4 \pm 3.4 \times 10^5$ for local commercial and $1.0 \times 10^5 \pm 2.5 \times 10^5$ for local home-made which were statistically significantly different ($P < 0.05$). Brands of ice cream of foreign commercial origin had significantly lower frequency of staphylococci, *E. coli* and lactose fermenters than those of local commercial and home-made sources. Coconut flavoured ice cream had the highest prevalence and mean counts of aerobic bacteria tested, while vanilla ice cream had the lowest one. The temperature and the pH of ice cream had a significant effect on its microbial load. Home-made ice cream in Trinidad appears to pose a high health risk to consumers compared to imported brands. It is imperative that bacteriological standards be enforced in order to prevent ice cream-borne gastroenteritis.

Key words: *Salmonella* spp., *Listeria* spp., *Escherichia coli*, staphylococci, contamination, temperature, pH.

Introduction

Ice cream, which is a milk product, is consumed globally and most consumption occurs outside homes and amongst children during the summer months in temperate countries and all year round in tropical nations^{30,39}. Although ice cream is a frozen product, it has been documented to be contaminated by bacterial pathogens such as *Salmonella* spp., *Listeria* spp., *Yersinia* spp., *Staphylococcus aureus*, *Escherichia coli*, coliforms, *Bacillus* spp. amongst others in many countries^{7, 11, 17, 25, 26, 39, 42, 44}. Contributing factors of ice cream contamination include poor sanitary practices during processing^{24, 33, 35, 36}, use of contaminated ingredients, particularly shell eggs^{9, 22, 33, 43}, contaminated scoop water⁴¹, post-process contamination^{24, 32, 35} and improper storage temperatures^{38, 39}.

The presence of pathogens in ice cream has been responsible for epidemics of salmonellosis, primarily due to *S. enteritidis*^{12, 16, 22} and listeriosis^{8, 19} elsewhere emphasizing the health risk posed to consumers by ice creams. To avoid ice cream-borne gastroenteritis, bacteriological standards have been established for ice cream by several governments^{27, 39}. In developing countries, although bacteriological standards may exist, there is always the problem of enforcement.

In Trinidad and Tobago, a tropical country with average temperatures over 30°C almost all year round, ice cream is very widely consumed. There has been an increase in the number of ice cream vendors, mostly selling home-made ice cream. In 1971, an epidemic of typhoid fever implicated home-made ice cream as

the vehicle³⁷. Adesiyun³ surveyed ice cream samples sold locally and detected *S. aureus* and *E. coli* but no *Salmonella* was recovered. To date, information is unavailable on the bacteriological quality of imported and locally processed ice cream. There are no reports on the factors that may affect its microbial quality under local conditions.

The study was, therefore, conducted to compare the bacteriological quality of ice cream originating from commercial foreign and local sources, as well as home-made products and to investigate the effect of brands, flavour, sale outlet, pH and temperature on its microbial load. The presence of O157 strain amongst isolates of *E. coli* was also determined.

Materials and Methods

Source of ice cream: The cross-sectional study design was used to sample ice cream from supermarkets and street vendors from commercial local and foreign manufacturers and from local home-made sources. During the visit to the sale outlets, the most common 3 brands were sampled.

Administration of questionnaire: At the sale outlet, a short questionnaire was administered to obtain information on the conditions of storage, transportation and handling of the product.

Sample collection: Twice weekly, outlets were visited for sample collection. At the supermarkets, which served as sources of commercial local and commercial foreign ice cream, samples were collected in the sealed containers in which they were sold. For home-made ice cream, usually sold in open containers, samples were purchased in styrofoam cups provided by the vendors.

Determination of pH and temperature of ice cream: The portable 704 Metrohm temperature-compensated pH meter (Metrohm Ltd., Switzerland) was used to determine the temperature and pH of ice cream samples immediately after purchase.

Isolation of *Listeria* spp.: A sterile spoon was used to scoop out ice cream from the containers of purchase into sterile plastic cups in the laboratory after which it was allowed to melt at room temperature. The FAO procedure was used for isolation. Briefly, for each sample, 25 g was weighed and added to 225 ml of *Listeria* enrichment broth (LEB) (Oxoid Ltd., Basingstoke, Hampshire, UK) and enriched for 48 h at 30°C. Enriched broth was then plated for isolation on *Listeria* selective agar (LSA) containing *Listeria* supplement (Oxoid Ltd., Basingstoke, Hampshire, UK) and incubated at 35°C for 24 h after which colonies with typical appearance of *Listeria*, greyish to black, were subcultured onto blood agar plates and thereafter subjected to biochemical tests¹⁸.

Isolation of *Salmonella* spp.: To detect *salmonella* in ice cream samples, the procedure described by the FAO was used. Briefly, 25 g of melted sample was pre-enriched in 225 ml of lactose broth (Oxoid Ltd., Basingstoke, Hampshire, UK) overnight at 37°C. As enrichment, 1 ml of each pre-enriched sample was then used to inoculate 9 ml each of selenite cysteine (SC) broth and tetrathionate (TT) broth and incubated overnight at 42°C and 37°C, respectively. Broth culture was then inoculated onto bismuth sulphite (BS) and xylose lysine desoxycholate (XLD) and incubated overnight at 37°C. *Salmonella* was identified as earlier recommended^{18, 28}. Suspect isolates were subjected to the slide agglutination test using the *salmonella* polyvalent antiserum A-I & Vi (Difco, Detroit, Michigan, USA).

Detection and enumeration of staphylococci: For qualitative detection of staphylococci, a loopful of molten ice cream was inoculated into Baird Parker agar (BPA) and incubated at 37°C for 48 h. Colonies that were black or greyish-black were Gram-stained and subjected to biochemical tests²⁸. For quantitative evaluation of staphylococci, 100-fold serial dilutions of molten ice cream in sterile saline were inoculated onto BPA plates and incubated at 37°C for 48 h. Typical colonies were enumerated using a Quebec Darkfield colony counter (Cambridge Instruments Inc., New York, USA), counts were expressed as colony-forming units (cfu) per g. The slide and tube coagulase tests²⁸ were used to detect *S. aureus* strains amongst staphylococcal isolates.

Detection and enumeration of *E. coli*: Ice cream samples were subcultured onto eosin methylene blue (EMB) plates (Oxoid Ltd., Basingstoke, Hampshire, UK) and incubated overnight at 37°C to detect the presence of *E. coli*. Colonies with greenish metallic sheen were subjected to biochemical test²⁸. To quantify the presence of *E. coli* in ice cream sample, 100-fold dilutions in sterile saline were plated on EMB plates and colonies with typical

appearance were enumerated. Counts were expressed as cfu per g of ice cream.

Detection of O157 strain of *E. coli*: Suspect isolates of *E. coli* were identified using standard methods and subsequently plated on sorbitol-McConkey agar (Oxoid Ltd., Basingstoke, Hampshire, UK). Inoculated plates were incubated at 37°C overnight and sorbitol fermenting (SF), which was reddish colonies, and non-sorbitol fermenting (NSF) colonies, including cream or colourless, were noted. All NSF isolates were plated on blood agar plates and incubated at 37°C overnight after which the *E. coli* O157 antiserum (Difco, Detroit, Michigan, USA) was used to detect O157 strain by slide agglutination test.

Determination of total aerobic plate count: The dilutions of ice cream used to enumerate staphylococci and *E. coli* in ice cream were similarly plated on nutrient agar plates and incubated overnight at 37°C. All colonies on plates with 30-300 colony forming units were counted. The counts were expressed as total aerobic plate count (TAPC) per g of ice cream.

Statistical analyses of data: The frequency of detection of various bacteria in ice cream samples were processed using the Statistical Package for Social Sciences (SPSS), version 10, and compared with chi-square tests. All statistical tests were performed 2-tailed and interpreted at an alpha of 0.05. The one-way ANOVA was used to compare the mean counts of the different types of bacteria, as well as the total aerobic plate counts in ice cream of various origins, flavours, brands, sale outlets, temperatures and pH.

Results

All 115 samples of ice cream tested were negative for *Salmonella* and *Listeria* spp. (Table 1). The TAPC per g of ice cream was highest in local home-made products ($1.0 \times 10^5 \pm 2.5 \times 10^5$) and lowest amongst foreign commercial products ($1.5 \times 10^2 \pm 2.9 \times 10^2$). The difference was statistically significant ($P < 0.05$). A similar pattern was observed for the prevalence and counts per g of ice cream for staphylococci, *E. coli*, lactose fermenters and non-lactose fermenters. All staphylococcal isolates were coagulase negative staphylococci (CNS), *i.e.* non-*S. aureus*.

Samples from all 14 brands of ice cream contained aerobic bacteria (Table 2). Of the 9 brands of commercial foreign sources, only 1 (11.1%) was positive for staphylococci, 1 (11.1%) for *E. coli* and 2 (22.2%) each for lactose fermenters and non-lactose fermenters. Samples of the 2 brands of local commercial and 3 of local home-made tested were positive for these bacteria.

Coconut flavoured ice cream, regardless of source, was most contaminated both in frequency and counts compared to the other 4 popular flavours tested (Table 3). The mean TAPC per g was $2.0 \times 10^5 \pm 5.8 \times 10^5$ for coconut ice cream compared with $9.0 \times 10^3 \pm 4.1 \times 10^4$ for vanilla ice cream. The frequency of detection of staphylococci, *E. coli*, lactose fermenters and non-lactose fermenters was 53.8% (7 of 13), 84.6% (11 of 13), 91.7% (11 of 12) and 83.3% (10 of 12), respectively, for coconut ice cream compared with 14.3% (4 of 28), 17.9% (5 of 28), 42.3% (11 of 26) and 30.8% (8 of 26) for vanilla flavoured ice cream.

The microbial load of ice cream samples from the 8 sale outlets studied varied from a TAPC per g of $2.7 \times 10^2 \pm 4.3 \times 10^2$ (Outlet E) to

$2.1 \times 10^5 \pm 6.6 \times 10^5$ (Outlet C) (Table 4). All samples collected from Outlet E were negative for *staphylococci* and only 10% (1 of 10) contained *E. coli*. A total of 120 isolates of *E. coli* were tested of which 29 (24.2%) were NSF but none was O157 strain.

The microbial load of ice cream decreased as its temperature decreased (Table 5). For ice cream samples with temperatures less than -8.8°C , the mean TAPC per g was $8.2 \times 10^3 \pm 3.2 \times 10^4$ compared with $1.1 \times 10^5 \pm 4.5 \times 10^5$ detected for samples with temperatures above -5.21°C . The frequency of isolation of aerobic bacteria across ice cream of varying temperatures was not significantly different ($p = 0.340$). The difference in the frequency of detection of *staphylococci* in ice cream samples across the temperature range investigated was, however, statistically significant ($p = 0.03$).

For the pH intervals (<6.463, 6.463-6.59, 6.58-6.71, 6.72-6.83 and >6.83) of ice cream investigated, regardless of source of ice cream, the pH had no statistically significant effect ($P > 0.05$) on the microbial load (TAPC per g) of samples studied. However, significantly higher frequency of detection of lactose fermenters ($p = 0.00$) and non-lactose fermenters ($p = 0.009$) was observed at low pH (less than 6.463), 81.0% (17 of 21 samples) than at comparatively high pH (over 6.83), 26.7% (4 of 15 samples) for lactose fermenters; while for non-lactose fermenters, at low pH, 66.7% (14 of 21) and at comparatively higher pH, 31.6% (6 of 19).

Discussion

It was significant that ice cream from foreign sources in Trinidad had better bacteriological quality than those that were either commercial or local home-made. This suggests that imported ice cream sold in the country, although comparatively more expensive than locally processed products, is safer for human consumption. A similar finding on the bacteriology of imported *versus* local product was reported for bottled water¹⁰ and raw meat¹ in the country.

It was no surprise that home-made ice cream, relatively inexpensive and popular locally, had a high microbial load which exceeded both local (Trinidad Public Health Laboratory, personal communication) and international standards^{27,39}. This is because there are no regulatory controls on the microbial loads of ice cream from these sources and therefore are susceptible to poor sanitary practices during processing and at the sale outlets. Furthermore, unlike the samples of commercial local and commercial foreign ice cream studied, which were all sold in packages, home-made ice cream were all sold open often by vendors in carts. The health risk posed to consumers by home-made ice cream in the country can therefore not be over-emphasized. Other investigators elsewhere have similarly reported that home-made ice cream has significantly higher level of bacterial contamination than commercial ice cream^{17,39,42}. Home-made ice cream has also been documented to have poor microbial quality elsewhere^{12,17,38,24,26} and epidemics of ice cream-borne gastroenteritis have been reported^{13,16,22,34}.

It is interesting to find that despite the high level of contamination of home-made ice cream, all samples tested, including those of commercial foreign and commercial local origin, were negative for *Salmonella* spp. and *Listeria* spp. Adesiyun³ also failed to detect *Salmonella* spp. from a total of 66 locally produced ice cream studied. This finding is in agreement with the reports of De Centorbi *et al.*¹⁴, Maifreni *et al.*²⁹, Aidara-Kane *et al.*⁷ and Warke *et al.*³⁹. It is, however, pertinent to mention that failure to detect both *Salmonella* and *Listeria* spp. from local ice cream does not make

it safe since other pathogens not assayed for in the current study may be present. In Trinidad and Tobago, both pathogens have been recovered from pre-processed bovine milk⁴. *Salmonella* and *Listeria* have been recovered from ice cream in other countries making them unfit for human consumption and in a number of cases were responsible for gastroenteritis^{11,15,17,21,26,42}.

The overall frequency of isolation of staphylococci (19.1%) found in this study is considerably lower than reported by Masud³¹ and Wouafo *et al.*⁴⁴. Home-made ice cream again had a significantly higher frequency of staphylococci (52.0%) compared with commercially processed (local and foreign) products. This could be attributed to poor handling during preparation and contamination from the environment during sale as reported by Wouafo *et al.*⁴⁴ and Kanbakan *et al.*²⁴. Lower frequencies of staphylococci and *S. aureus* have been also reported for ice cream by Wouafo *et al.*⁴⁴, Kanbakan *et al.*²⁴ and El-Sharef *et al.*¹⁷.

Regarding health concern, 88% of home-made ice cream sampled were positive for *E. coli*, with as high as 96% contaminated by enteric bacteria. This frequency is much higher than 51.4% and 26.7% of home-made and commercial ice cream, respectively, reported to be positive for *E. coli*⁴². Comparatively, lower frequency of isolation of *E. coli* from ice cream has been reported for other countries^{17,31}.

Coliforms and *E. coli* may contaminate ice cream during processing through contaminated water or faecal material as a result of poor sanitary practices. Pipeborne treated water processed by the nation's Water and Sewerage Authority (WASA) was reported to have unacceptable coliform level in water supply to urban and rural communities in Trinidad^{6,40}. It was also demonstrated that containers or tanks used to store water at homes contributed to coliform contamination of water⁴⁰. There is, therefore, a possibility that the water used during the production and sale of ice cream, particularly home-made one, may have served as a vehicle for *E. coli* and other coliforms to contaminate the ice cream studied. It is relevant to mention that at the sale outlet, particularly when sold in open containers which require dispensing of the product for sale, scoop water may serve as vehicle of coliform contamination of ice cream as reported by Wouafo *et al.*⁴⁴, Wilson *et al.*⁴¹ and Kanbakan *et al.*²⁴.

Strain O157 was not detected amongst all isolates of *E. coli* studied, an indication that ice cream sold in Trinidad is not expected to be an important source of gastroenteritis due to verocytotoxigenic *E. coli* (VTEC). However, El-Sharef *et al.*¹⁷ reported that 1.3% of home-made ice cream sold in Libya was contaminated by *E. coli* O157. Milk-borne, including ice cream, gastroenteritis due to VTEC strains are well documented in the literature^{20,23,34}. An earlier study in Trinidad recovered VTEC strains from pre-processed bovine milk^{2,5}.

Brands and sale outlets did not have a significant effect on the ice cream samples *per se*, as the source (home-made, local commercial or foreign commercial) appeared to be the most important factor influencing the microbial quality of the product. It was observed that all nine brands of ice cream from foreign sources had both low frequency and mean counts of bacteria tested, while the frequency and counts of all brands from local sources (commercial local and home-made) were high.

In the country, vanilla ice cream, unquestionably the most popular flavour, had a better bacteriological quality than coconut flavoured ice cream which had the poorest quality (frequency and

Table 1. Frequency and counts of bacteria in ice cream by source of manufacture.

Type of ice cream	No. of samples ^a tested	Mean \pm SD ^b TAPC, cfu/g	Frequency and counts of bacteria							
			Staphylococci		<i>E. coli</i>		Lactose-fermenters ^c		Non-lactose fermenters ^c	
			No. (%) of samples positive	Mean \pm SD count (cfu/g)	No. (%) of samples positive	Mean \pm SD count (cfu/g)	No. (%) of samples positive	Mean \pm SD count (cfu/g)	No. (%) of samples positive	Mean \pm SD count (cfu/g)
Foreign commercial	52	1.5x10 ² \pm 2.9x10 ²	1 (1.9)	1.9x10 ³ \pm 1.4x10 ⁰	2 (3.8)	3.3x10 ⁰ \pm 2.1x10 ¹	8 (17.0)	2.1x10 ¹ \pm 6.9 x 10 ¹	8 (17.0)	2.4x10 ¹ \pm 9.4x10 ¹
Local commercial	38	7.4x10 ⁴ \pm 3.4x10 ⁵	8 (21.1)	9.5x10 ³ \pm 5.6x10 ⁴	10 (26.3)	2.1x10 ² \pm 8.8x10 ²	30 (85.7)	8.2x10 ² \pm 1.9x10 ³	25 (71.4)	1.4x10 ² \pm 3.7x10 ²
Local home-made	25	1.0x10 ⁵ \pm 2.5x10 ⁵	13 (52.0)	2.9x10 ² \pm 7.0x10 ²	22 (88.0)	5.5x10 ² \pm 2.0x10 ³	24 (100.0)	1.6x10 ² \pm 4.0 x 10 ³	23 (100.0)	3.5x10 ³ \pm 1.0x10 ⁴
Total	115	4.6x10 ⁵ \pm 2.3x10 ⁵	22 (19.1)	3.1x10 ³ \pm 3.3x10 ⁴	34 (29.6)	1.9x10 ² \pm 1.1x10 ³	106 ^f (58.5)	6.4x10 ² \pm 2.2x10 ³	56 (53.3)	8.3x10 ² \pm 5.9x10 ³

^aAll samples were negative for *Salmonella* and *Listeria* spp.^bTotal aerobic plate count and all samples were positive for aerobic bacteria.^cNo. of samples tested for lactose fermenters and non-lactose fermenters.^dTwenty-three samples were tested for non-lactose fermenters.^eLactose fermentation and non-lactose fermentation on McConkey agar.**Table 2.** Frequency of isolation of bacteria in ice cream by brands.

Brand of ice cream ^a	No. of samples tested ^c	No. (%) positive ^b for		No. of samples tested ^d	No. (%) positive ^b for	
		Aerobic bacteria	<i>Staphylococci</i>		<i>E. coli</i>	Lactose fermenter
A ₁	16	16 (100.0)	0 (0.0)	14	0 (0.0)	0 (0.0)
A ₂	5	5 (100.0)	0 (0.0)	12	0 (0.0)	0 (0.0)
A ₃	4	3 (75.0)	0 (0.0)	3	0 (0.0)	0 (0.0)
A ₄	2	1 (50.0)	0 (0.0)	1	0 (0.0)	0 (0.0)
A ₅	8	7 (87.5)	1 (12.5)	8	1 (12.5)	3 (37.5)
A ₆	7	7 (100.0)	0 (0.0)	7	2 (28.6)	4 (57.1)
A ₇	6	5 (83.3)	0 (0.0)	6	0 (0.0)	0 (0.0)
A ₈	2	1 (50.0)	0 (0.0)	2	0 (0.0)	0 (0.0)
A ₉	2	2 (100.0)	0 (0.0)	2	0 (0.0)	0 (0.0)
B ₁	12	12 (100.0)	5 (41.7)	11	3 (25.0)	10 (90.9)
B ₂	26	26 (100.0)	3 (11.5)	24	6 (23.1)	15 (62.5)
C ₁	12	12 (100.0)	7 (58.3)	11	11 (100.0)	10 (100.0) ^e
C ₂	8	8 (100.0)	3 (37.5)	8	8 (100.0)	8 (100.0)
C ₃	5	5 (100.0)	3 (60.0)	5	3 (60.0)	5 (100.0)

^aA₁-A₉ are foreign commercial, B₁-B₂ are local commercial^bNo. of samples tested for aerobic bacteria, staphylococci and *E. coli*.^cNo. of samples tested for lactose fermenters and non-lactose fermenters.^dTen samples were tested for non-lactose fermenters.^eAll samples were negative for *Salmonella* and *Listeria* spp.

Table 3. Frequency and counts of bacteria in ice cream by popular flavours ^a.

Flavour	No. of samples tested ^a	Aerobic bacteria		Staphylococci		<i>E. coli</i>		Lactose fermenters		Non-lactose fermenters	
		Mean ± SD TAPC ^b , cfu/g	No. (%) of samples positive	Mean ± SD count (cfu/g)	No. (%) of samples positive	Mean ± SD count (cfu/g)	No. (%) of samples positive	No. (%) of samples positive	Mean ± SD count (cfu/g)	No. (%) of samples positive	Mean ± SD count (cfu/g)
Vanilla	28	9.0x10 ³ ± 4.1x10 ⁴	4 (14.3)	7.8x10 ⁰ ± 2.0x10 ¹	5 (17.9)	2.0x10 ¹ ± 6.3x10 ¹	11 (42.3)	3.7x10 ² ± 9.5x10 ²	8 (30.8)	1.9x10 ² ± 7.9x10 ²	
Chocolate	17	1.1x10 ⁴ ± 3.9x10 ⁴	3 (17.6)	7.6x10 ⁰ ± 1.9x10 ¹	3 (17.6)	2.1x10 ² ± 6.9x10 ¹	7 (46.7)	3.7x10 ² ± 8.0x10 ²	7 (46.7)	1.2x10 ² ± 2.7x10 ²	
Coconut	13	2.0x10 ⁵ ± 5.8x10 ⁵	7 (53.8)	2.7x10 ⁴ ± 9.7x10 ⁴	11 (84.6)	8.7x10 ² ± 2.7x10 ³	11 (91.7)	2.4x10 ³ ± 5.6x10 ³	10 (83.3)	5.4x10 ³ ± 1.7x10 ⁴	
Strawberry	6	1.9x10 ⁵ ± 4.5x10 ⁵	0 (0.0)	NA	3 (50.0)	4.2x10 ¹ ± 8.3x10 ¹	4 (66.7)	2.8x10 ² ± 3.9x10 ²	4 (66.7)	2.8x10 ² ± 3.1x10 ²	
Rum and raisin	5	1.6x10 ⁴ ± 2.7x10 ⁴	1 (20.0)	2.0x10 ⁰ ± 4.5x10 ⁰	1 (20.0)	5.0x10 ¹ ± 1.1x10 ²	3 (60.0)	3.5x10 ² ± 6.0x10 ²	3 (60.0)	4.2x10 ² ± 8.3x10 ²	
Others ^e	46	2.4x10 ⁴ ± 9.0x10 ⁴	7 (15.2)	4.4x10 ¹ ± 2.5x10 ²	11 (23.9)	1.3x10 ² ± 6.9x10 ²	26 (61.9)	5.1x10 ² ± 1.6x10 ³	24 (58.5) ^d	3.0x10 ² ± 5.5x10 ²	

^aNo. of samples tested for aerobic bacteria, staphylococci and *E. coli*. ^bAll samples contained aerobic bacteria. ^cNo. of samples tested for lactose fermenters and non-lactose fermenters. ^dForty-one samples were tested for non-lactose fermenters. ^eOthers included: Cookies n' Cream (5 samples), Dulce de Leche (5 samples), Sourtop (5 samples), Peanut (4 samples), RockyRoad (3 samples), Cherry Vanilla (3 samples) and seven others with one sample each.

Table 4. Microbial load of ice cream by outlet of purchase.

Outlet	No. of samples tested ^a	Aerobic bacteria		Staphylococci		<i>E. coli</i>		Lactose fermenters		Non-Lactose fermenters	
		No. (%) positive	Mean ± SD TAPC, cfu/g	No. (%) positive	Mean ± SD, count (cfu/g)	No. (%) positive	Mean ± SD, count (cfu/g)	No. (%) positive	Mean ± SD, count (cfu/g)	No. (%) positive	Mean ± SD, count (cfu/g)
A	40	37 (92.5)	4.4x10 ⁴ ± 1.8x10 ⁵	5 (12.5)	9.3x10 ⁰ ± 2.9x10 ¹	17 (42.5)	4.3x10 ³ ± 1.7x10 ³	22 (55.0)	1.0x10 ³ ± 3.5x10 ³	20 (50.0)	1.7x10 ³ ± 9.5x10 ³
B	30	29 (96.7)	1.0x10 ⁴ ± 4.0x10 ⁴	7 (23.3)	8.6x10 ¹ ± 3.3x10 ²	6 (20.0)	4.0x10 ¹ ± 1.3x10 ²	9 (42.9)	1.9x10 ² ± 3.8x10 ²	8 (40.0) ^c	3.8x10 ² ± 9.9x10 ²
C	10	10 (100.0)	2.1x10 ⁵ ± 6.6x10 ⁵	3 (30.0)	3.5x10 ⁴ ± 1.1x10 ⁵	3 (30.0)	1.2x10 ¹ ± 2.2x10 ¹	5 (50.0)	7.8x10 ² ± 1.4x10 ³	3 (30.0)	5.4x10 ¹ ± 8.7x10 ¹
D	10	10 (100.0)	6.2x10 ² ± 8.2x10 ²	1 (10.0)	1.0x10 ⁰ ± 3.2x10 ⁰	2 (20.0)	1.6x10 ¹ ± 4.7x10 ¹	6 (60.0)	1.9x10 ² ± 2.5x10 ²	5 (50.0)	1.9x10 ¹ ± 2.5x10 ¹
E	10	9 (90.0)	2.7x10 ² ± 4.3x10 ²	0 (0.0)	NA	1 (10.0)	3.0x10 ⁰ ± 9.5x10 ⁰	5 (50.0)	1.1x10 ² ± 1.6x10 ²	8 (80.0)	1.9x10 ² ± 9.3x10 ²
F	5	5 (100.0)	1.5x10 ⁵ ± 2.5x10 ⁵	3 (60.0)	6.4x10 ² ± 1.4x10 ³	3 (60.0)	6.2x10 ¹ ± 8.4x10 ¹	5 (100)	9.4x10 ² ± 1.4x10 ³	5 (100.0)	1.5x10 ³ ± 8.4x10 ³
G	5	5 (100.0)	2.7x10 ³ ± 8.0x10 ³	1 (20.0)	2.0x10 ¹ ± 4.5x10 ¹	1 (20.0)	2.0x10 ¹ ± 4.5x10 ¹	5 (100)	4.0x10 ² ± 3.1x10 ²	3 (60.0)	8.0x10 ¹ ± 7.8x10 ²
H	5	5 (100.0)	5.3x10 ⁴ ± 1.3x10 ⁵	2 (40.0)	2.4x10 ¹ ± 3.4x10 ¹	1 (20.0)	5.6x10 ² ± 1.3x10 ³	5 (100)	1.3x10 ³ ± 1.4x10 ³	4 (80.0)	2.7x10 ² ± 3.2x10 ²

^aNo. of samples tested for aerobic bacteria, staphylococci and *E. coli*. ^bNo. of samples tested for lactose fermenters and non-lactose fermenters. ^cTwenty samples were tested for lactose fermenters.

Table 5. Microbial load of ice cream by temperature of product on purchase.

Range of temperature (°C)	No. of samples tested ^a	Mean ± SD counts, (cfu/g) of:					
		Aerobic bacteria	Staphylococci	<i>E. coli</i>	No of samples tested ^b	Lactose fermenters	Non-lactose fermenters
>-5.21	22	1.1x10 ⁵ ± 4.5x10 ⁵	1.6x10 ⁴ ± 7.5x10 ⁴	2.4x10 ³ ± 1.0x10 ²	22	3.2x10 ² ± 6.4x10 ²	8.5x10 ² ± 2.2x10 ³
-5.22 to -6.3	20	2.8x10 ⁴ ± 7.4x10 ⁴	2.9x10 ¹ ± 4.7x10 ¹	5.7x10 ² ± 2.2x10 ³	19	3.5x10 ³ ± 1.4x10 ⁴	1.6x10 ³ ± 4.6x10 ³
-6.4 to -7.45	24	7.2x10 ⁴ ± 2.5x10 ⁵	4.2x10 ¹ ± 2.0x10 ⁰	3.9x10 ¹ ± 8.5x10 ¹	23	2.5x10 ² ± 6.1x10 ²	4.5x10 ² ± 9.6x10 ²
-7.46 to -8.7	20	1.5x10 ⁴ ± 4.0x10 ⁴	1.0x10 ⁰ ± 3.1x10 ⁰	2.4x10 ¹ ± 6.1x10 ¹	19	1.3x10 ² ± 3.1x10 ²	1.4x10 ² ± 2.0x10 ²
<-8.8	24	8.2x10 ³ ± 3.2x10 ⁴	5.8x10 ⁰ ± 2.2x10 ¹	1.3x10 ² ± 5.7x10 ²	22	4.6x10 ³ ± 2.1x10 ³	2.6x10 ² ± 8.5x10 ²

^aNo. of samples tested for aerobic bacteria, staphylococci and *E. coli*.

^bNo. of samples tested for lactose fermenters and non-lactose fermenters.

count) of all the flavours studied. This finding is at variance with the report of Maifreni *et al.*²⁹ where no statistically significant difference was detected in the means of TAPC, coliforms and yeast counts in various flavours of ice cream studied. It is known that ingredients used in the preparation of ice cream are potential sources of contamination^{22, 33, 35}.

It is worthy to note that the temperature of ice cream had no significant effect on the mean TAPC per g, which was an unexpected finding. High temperature or fluctuating temperatures during storage of ice cream, particularly those sold in open containers and in high ambient temperature, is known to facilitate the multiplication of bacteria^{17, 39}. Warke *et al.*³⁹ reported that in 10 days of frozen storage, *L. monocytogenes* counts grew to >1 log and 1 log cycle at 8-10°C and 2-4°C, respectively.

The normal range of the pH of ice cream is close to neutral, 6-7²⁴, all the samples of ice cream in the current study had pH values within this pH range. Therefore, it was not a surprise that the pH of the ice cream studied did not significantly affect its microbial load. The finding that lactose fermenting bacteria were significantly more recovered from ice cream with pH of <6.46 than those with pH of >6.83 was expected. It may be explained in part by the fact that most coliforms are lactose fermenters, which may, by their activity, lower the pH of ice cream or they are more adapted to survive in lower pH.

Conclusions

It was concluded that home-made ice cream sold in Trinidad and Tobago pose the greatest health risk to consumers because of their poor bacteriological quality. It is imperative that the authorities initiate steps to ensure quality control of the product to which the consumers are exposed. It is only, by doing so, that the risk posed by the product to the public will be reduced.

References

- Adesiyun, A. A. 1993. Prevalence of *Listeria* spp., *Campylobacter* spp., *Salmonella* spp., *Yersinia* spp. and toxigenic *Escherichia coli* on meat in Trinidad. *Food Microbiol.* **10**:395-403.
- Adesiyun, A. A. 1994. Bacteriological quality and associated public health risk of pre-processed bovine milk in Trinidad. *Int. J. Food Microbiol.* **21**:253-261.
- Adesiyun, A. A. 1995. Bacteriologic quality of some Trinidadian ready-to-consume foods or drinks and possible health risks. *J. Food Prot.* **58**:651-655.

- Adesiyun, A. A., Webb, L. A., Romain, H. and Kaminjolo, J. S. 1996. Prevalence of *Salmonella* spp., *Listeria monocytogenes*, *Campylobacter* spp., *Yersinia enterocolitica* and *Cryptosporidium* in bulk milk, cows' faeces and effluents of dairy farms in Trinidad. *Rev. Elev. Med. Vet. Pays Trop.* **49**:303-309.
- Adesiyun, A. A., Webb, L. A., Romain, H. and Kaminjolo, J. S. 1997. Prevalence and characteristics of *Escherichia coli* strains isolated from milk and faeces of cows on dairy farms in Trinidad. *J. Food Prot.* **60**:1174-1181.
- Agard, L., Alexander, C., Green, S., Jackson, M., Patel, S. and Adesiyun, A. A. 2002. Microbial quality of water supply to an urban community in Trinidad. *J. Food Prot.* **65**:1297-1303.
- Aidara-Kane, A., Ranaivo, A., Spiegel, A., Catteau, M. and Rocourt, J. 2000. Microbiological quality of street-vendor ice cream in Dakar. *Dakar Med.* **45**:20-24.
- Anon. 1987. FDA, CDC, investigate ice cream link to listeriosis outbreak. *Food Chem. News* **29**:3.
- Anon. 1990. Vanilla ice cream recalled. FDA Enf. Report 7 November.
- Bharath, J., Mosodeen, M., Motilal, S., Sandy, S., Sharma, S., Tessaro, T., Thomas, K., Umamaheswaran, M., Simeon, D. and Adesiyun, A. A. 2002. Microbial quality of domestic and imported brands of bottled water in Trinidad. *Int. J. Food Microbiol.* **81**:53-62.
- Cardano, A. M. and Rocourt, J. 2001. Occurrence of *Listeria monocytogenes* in food in Chile. *Int. J. Food Microbiol.* **70**:175-178.
- Centers for Disease Control. 1994. Outbreak of *Salmonella enteritidis* associated with home-made ice cream. *MMWR, Florida* **43**:669-671.
- Daniels, N. A., McKinnon, L., Rowe, S. M., Bean, N. H., Griffin, P. M. and Mead, P. S. 2002. Foodborne disease outbreaks in United States schools. *Pediatr. Infect. Dis. J.* **21**:623-628.
- De Centorbi, O. P., De Guzman, A. C., De Cuadrado, A. A., Laciari, A. L., Alcaraz, L. E., De Malan, M. C. and De Pederiva, N. B. 1989. Determination of the sanitary quality and detection of *Salmonella* spp. and *Yersinia enterocolitica* in ice cream. *Rev. Argent. Microbiol.* **21**:63-69.
- De Tamsut, L. S. and García, C. E. 1989. Microbiological quality of vanilla ice cream manufactured in Caracas, Venezuela. *Arch. Latinoam. Nutr.* **39**:46-56.
- Dodhia, H., Kearney, J. and Wartburton, F. 1998. A birthday party, home-made ice cream and an outbreak of *Salmonella enteritidis* phage type 6 infection. *Commun. Dis. Publ. Health* **1**:31-34.
- El-Sharef, N., Ghenghesh, K. S., Abognah, Y. S., Gnan, S. O. and Rahouma, A. 2005. Bacteriological quality of ice cream in Tripoli, Libya. *Food Control* **17**:637-641.
- Food and Agricultural Organization (FAO) 1992. Manual of Food Quality Control. 4, Rev. 1. Microbiological analysis. FAO, Rome, pp. 24-48.
- Farrag, S. A. and Marth, E. H. 1992. Interactions between *Listeria monocytogenes* and other psychotropic bacteria in dairy foods: A review.

- Food Aust. **44**:284-286.
- ²⁰Gray, L. D. 1995. *Escherichia*, *Salmonella*, *Shigella* and *Yersinia*. In Murray, P. R., Baron, E. J., Pfaller, M. A., Tenover, F. C. and Tenover, R. H. (eds). Manual of Clinical Microbiology. ASM Press, Washington, pp. 450-456.
- ²¹Greenwood, M. H., Roberts, D. and Burden, P. 1991. The occurrence of *Listeria* species in milk and dairy products: A national survey in England and Wales. *Int. J. Food Microbiol.* **12**:197-206.
- ²²Hennessy, T. W., Hedberg, C., Slutsker, L., White, K. E., Besser-Wiek, J. M., Moen, M. E., Feldman, J., Coleman, W. W., Edmonson, L. M., McDonald, K. L. and Osterholm, M. T. 1996. A national outbreak of *Salmonella enteritidis* infection from ice cream. *N. Engl. J. Med.* **334**:1281-1286.
- ²³Karmali, M. A. 1989. Infection by verocytotoxin-producing *Escherichia coli*. *Clin. Microbiol. Rev.* **2**:15-38.
- ²⁴Kanbakan, U., Con, A. H. and Ayar, A. 2004. Determination of microbiological contamination sources during ice cream production in Denizli, Turkey. *Food Control* **15**:463-470.
- ²⁵Kristufkova, Z. and Simkovicova, M. 1988. An epidemic of enterotoxigenic *Staphylococcus aureus* from ice cream containing staphylococcal TSST-1. *Ceskosl. Epidemiol. Mikrobiol. Immunol.* **37**:36-38.
- ²⁶Kruij, S. L., Soares, J. L., Ping, S. and Sainte-Marie, F. F. 2001. Microbiological quality "ice, ice cream sorbet" sold on the streets of Phnom Penh. April 1996-April 1997. *Bull. Soc. Pathol. Exot.* **94**:411-414.
- ²⁷Libyan Standard No. 364 for Ice Cream (LSIC) 1997. General Secretary for Planning and Economics, Tripoli, Libyan Arab Jamahiriya.
- ²⁸McFaddin, J. F. 2000. Biochemical Tests for Identification of Medical Bacteria. Williams and Wilkins, New York, USA.
- ²⁹Maifreni, M., Civilini, M., Domenis, C., Manzano, M., Di Prima, R. and Comi, G. 1993. Microbiological quality of artisanal ice cream. *Int. J. Hyg. Environ. Med.* **194**:553-570.
- ³⁰Mann, E. J. 1988. Ice cream. Part I. *Dairy Int.* **53**:13-14.
- ³¹Masud, T. 1989. Microbiological quality and public health significance of ice cream. *J. Pakistan Med. Assoc.* **39**:102-104.
- ³²Microbiological Risk Assessment of Ice Cream. 2001. An Evaluation of Microbiological Surveillance of Ice Cream. Report No. 7. Risk Assessment Studies, Risk Assessment Section, Food and Environmental Hygiene Department, Hong Kong.
- ³³Morgan, D., Mawer, S. L. and Harman, P. L. 1994. The role of home-made ice cream as a vehicle of *Salmonella enteritidis* phage type 4 infection from fresh shell eggs. *Epidemiol. Infect.* **113**:21-29.
- ³⁴Payne, C. J., Petrovic, M., Roberts, R. J., Paul, A., Linnane, E., Walker, M., Kitby, D., Burgess, A., Smith, R. M., Cheasty, T., Willshaw, G. and Salmon, R. L. 2003. Verocytotoxin-producing *Escherichia coli* O157 gastroenteritis in farm visitors, North Wales. *Emerg. Infect. Dis.* **9**:526-530.
- ³⁵Reij, M. W. and Den Aantrekker, E. D. 2004. ILSI Europe Risk Analysis in Microbiology Task Force. Recontamination as a source of pathogens in processed foods. *Int. J. Food Microbiol.* **91**:1-11.
- ³⁶Rodríguez-Álvarez, C., Hardisso, A., Álvarez, R., Arias, A. and Sierra, A. 1995. Hygienic-sanitary indicators of ice cream sold at the retail outlets. *Acta Alimentaria* **20**:69-80.
- ³⁷Taylor, A. Jr., Santiago, A., González-Cortes, A. and Gangarosa, E. J. 1974. Outbreak of typhoid fever in Trinidad in 1971 traced to a commercial ice cream product. *Amer. J. Epidemiol.* **100**:150-157.
- ³⁸Walker, S. J., Archer, P. and Banks, J. G. 1990. Growth of *Listeria monocytogenes* at refrigeration temperatures. *J. Appl. Bacteriol.* **68**:157-162.
- ³⁹Warke, R., Kamat, A., Kamat, M. and Thomas, P. 2000. Incidence of pathogenic psychrotrophs in ice creams sold in some retail outlets in Mumbai, India. *Food Control* **11**:77-83.
- ⁴⁰Welch, P., David, J., Clarke, W., Trinidad, A., Penner, D., Berstein, S., McDougall, L. and Adesiyun, A. A. 2000. Microbial quality of water in rural communities in Trinidad. *Pan. Amer. J. Public Health* **83**:172-179.
- ⁴¹Wilson, I. G., Heaney, J. C. and Weatherup, S. T. 1997. The effect of ice-cream-scoop water on the hygiene of ice creams. *Epidemiol. Infect.* **119**:35-40.
- ⁴²Windrantz, P. and Arias, M. L. 2000. Evaluation of the bacteriological quality of ice cream sold at San Jose, Costa Rica. *Arch. Latinoam. Nutr.* **50**:301-303.
- ⁴³World Health Organization (WHO) 1988. Food borne Listeriosis. World Health Organization Report of Informal Working Group on Listeriosis, WHO/EHE/FOS 88.5, Geneva.
- ⁴⁴Wouafo, M. N., Njine, T. and Tailliez, R. 1996. Hygienic and microbiologic quality of ice creams produced in Cameroon: A public health problem. *Bull. Soc. Pathol. Exot.* **89**:358-362.