RESULTS AND DISCUSSION

Part I

Properties of Commercial Flavoured Frozen Yoghurt

INTRODUCTION

Frozen yoghurt goes back a long way in history, but only more recently has it become a viable commercial product in Egyptian market. However, there are no specification for the products in regard to the chemical, physical properties and type of strains used and storage conditions.

The chemical composition, physical properties and more important, the microbiological quality of frozen yoghurt sold to the consumer are of great important.

The study of the chemical composition and microbiological quality is expected to show if the samples are within the legal standards. But no CODEX, Egyptian or Federal Standards of identity exist for frozen yoghurt at present, consequently the composition of commercialized frozen yoghurt varies greatly across brands. However, a proposed Federal Standards (USA) were submitted by the International Ice Cream Association to the FDA in 1989 (Mitten, 1989 and Guinard *et al.*, 1994). Moreover, a proposed Egyptian Standards are discussed by the Technical Committee on Milk and Milk Products in Egyptian Organization For Standardization and Quality Control (ESO).

Despite a general consensus that frozen yoghurt is fresh yoghurt, stabilized, whipped and frozen similarly to ice cream, little information is available on the actual nature of the commercial product. Most frozen yoghurt manufacturers resort to use of cows', buffaloes' milk composite milk products and/or reconstituted and recombined milk. Therefore, basic formulas may have variations and modifications as demanded by the ingredient available, consumer buying habits, competitions, costs and finished product quality expected. Consequently the chemical composition of frozen yoghurt in the market is expected to vary greatly. However, milk fat, protein, total solids and ash are the main determinants of nutritional value and textural quality consequently, physical properties (i.e. melting resistance). Total carbohydrate, reducing sugars, acid and acetaldehyde are the main contributers to flavour.

Regarding microbiological quality, if frozen yoghurt is manufactured, handled or stored under unfavourable conditions, some contaminants may gain entrance and grow and hence frozen yoghurt becomes a healthy hazard rather than being a nutritious product. These also may cause spoilage of the product upon extended storage.

Consequently, the study of some of the nuisance groups of microorganisms as coliform group, *E. coli* and yeasts, moulds in frozen yoghurt is expected to through some light on the hygienic conditions of its manufacture and handling as well as the expected defects upon storage and to its health fullness to the consumers.

In addition, the counts of viable lactic acid bacteria have also some benefits on the health aspects of frozen yoghurt.

Consequently, the objective of the present part was to collect and analyze commercial frozen yoghurt from Cairo and Giza regions for their composition, microbial numbers, acidity, physical and flavour characteristics.

EXPERIMENTAL PROCEDURES

Twenty five samples of commercial flavoured frozen yoghurt, representing 5 brands on the Cairo and Giza markets (Egypt), were collected at random throughout the summer of 1995, 9 of these samples were labelled as vanilla frozen yoghurt, 4 as strawberry frozen yoghurt, 4 as chocolate frozen yoghurt, 4 as coffee frozen yoghurt and the remaining samples (4) were labelled as mango frozen yoghurt. Upon receipt at the dairy laboratory, the samples were placed at -18 ± 2°C until ready for use. Then they were thawed in approximately 4 h at room temperature and analyzed in duplicate. All samples were analyzed chemically for titratable acidity, pH, fat, total solids, protein, total carbohydrate, reducing sugars, and acetaldehyde. Melting resistance was determined. The lactic acid bacterial counts for *St. salivarius* ssp. *thermophilus* and *Lb. delbrueckii* ssp. *bulgaricus*; coliform group; *E. coli*; and yeast and mould counts were also carried out. The results of the survey obtained in this study were statistically analyzed. However, all analysis were undertaken as mentioned under "material and methods"

RESULTS AND DISCUSSION

I- Titratable acidity (T.A.) and pH values:

The titratable acidity of 25 commercial flavoured frozen yoghurts represented by 5 Brands ranged from 0.04 to 0.63% (averaged 0.31%), and the lowest pH was 5.85, while the highest was 7.88 with an average of 6.95 (Table, 3). But pH values of strawberry and mango frozen yoghurts examined were found to be higher than that mentioned by Olsen (1990) who mentioned that, products containing fruit flavourings have a pH of 4.4 to 5.7 to emphasis a fresh fruit taste. The higher pH values in frozen yoghurts examined might be attributed to the use of pH controlling agents. Invariably, the different Brands established a characteristic titratable acidity and pH values range independent of flavour employed. For example, vanilla frozen yoghurts displayed a titratable acidity between 0.04 and 0.63% for its samples and pH values of 5.93 to 7.72. Mango frozen yoghurts had a titratable acidity between 0.04 to 0.05% and a pH values between 7.13 to 7.76 (Table, 3). These findings may be due to the use of pH contolling againsts.

These results are partially in agreement with those obtained by Kosikowski (1981) who found that, the T.A. of 34 commercial flavoured frozen yoghurt represented by 11 Brands ranged from 0.31to 1.35% and displayed a pH values between 4 and 6.5; Brown *et al.* (1991) who found that, 97 samples of frozen hard and soft frozen yoghurt representing 24 Brands showed 0.09 to 0.71% for T.A. and pH values ranged between 4.75 and 7.03.; and Whitehead *et al.* (1993) who found that, T.A. were >0.4% for 20 fruit flavoured frozen yoghurt; and only 43% of the non-fruit-flavoured products had T.A. >0.3%.

Table (4) and Fig. (2) show the frequency distribution of all collected samples. It clearly show that the majority of samples (64%) had low T.A. that lies within the range of 0.04 to 0.47%, which are illegal according to the T.A. which represent general accepted industry Standards (min. 0.5%) (Arbuckle, 1986). On the other hand, 13 out of 25 samples (52%) had a particularly legal T.A. values (0.39 to 0.63%) according to the proposed Federal Standards for T.A. (min. 0.3%).

The samples of low T.A. and high pH values might be attributed to the use of pH controlling agents with frozen yoghurt mixes, or to an alternative practice might be simply that of adding a bacterial cultures and then not incubating the inoculated mixes.

Table (3): Chemical properties of frozen yoghurt collected from some local markets.

Sample No.	Brands	T.A. %	pH	Fat %	T.S. %	Protein %	Ash %	Total carbo. %	Red. sugar %	acetaldehyde ppm
1		0.12	7.33	4.42	34.25	3.60	0.71	21.45	5.97	
2		0.08	7.60	4.58	32.87	4.52	0.74	23.63	4.27	2
3		0.04	7.72	4.84	33.94	3.90	0.83	22.66	4.01	
4		0.08	7.56	4.63	34.44	3.85	0.78	26.50	3.86	
5	Vanilla	0.63	5.93	4.33	31.32	3.53	0.93	21.81	6.85	
6		0.61	6.20	4.90	34.60	3.95	0.91	22.44	7.80	
7		0.58	6.40	4.17	38.70	4.11	0.88	21.63	8.24	12
8		0.58	5.97	4.88	33.60	3.96	1.04	19.20	7.25	
9		0.57	5.96	5.35	33.40	3.66	0.97	21.76	8.47	
10		0.47	5.85	4.26	32.98	4.40	0.78	21.75	7.58	
11	Straw-	0.39	5.95	4.61	35.24	3.80	0.76	21.70	8.25	
12	berry	0.44	5.88	4.68	34.85	3.60	0.74	22.59	9.15	22
13		0.50	6.02	4.67	34.60	3.70	0.81	21.30	7.02	
14		0.50	7.88	3.76	35.27	4.20	0.83	23.36	8.79	
15	Choco-	0.50	7.81	4.80	34.21	4.00	0.79	23.74	7.30	18
16	late	0.45	7.80	4.19	35.67	3.86	0.73	22.50	7.65	
17		0.50	7.82	4.83	32.44	3.96	1.01	21.70	5.67	
18		0.15	7.09	4.62	32.35	3.70	0.82	23.50	5.90	
19	Coffee	0.16	7.02	4.97	34.57	3.93	0.69	22.80	7.30	2
20		0.15	7.05	4.82	34.93	3.79	0.75	23.02	8.44	
21		0.15	7.30	4.75	34.60	3.87	0.71	20.57	8.13	
22		0.04	7.43	3.98	33.93	3.74	0.83	21.13	6.12	
23	Mango	0.05	7.13	4.21	37.87	3.91	0.91	23.30	8.43	33
24		0.05	7.76	4.60	34.46	4.01	0.88	21.60	7.13	
25		0.05	7.53	4.50	34.25	3.81	0.86	23.20	7.35	
Average		**0.31**	**6.95**	**4.57**	**35.74**	**3.89**	**0.83**	**22.35**	**7.11**	

Table (4): Frequency distribution of titratable acidity of frozen yoghurts collected from some local markets .

No.	Class intervals	Frequency	
		No. of samples	%
1	0.04-0.12	8	32
2	0.13-0.21	4	16
3	0.22-0.30	-	-
4	0.31-0.39	1	4
5	0.40-0.48	3	12
6	0.49-0.57	4	16
7	0.58-0.66	5	20

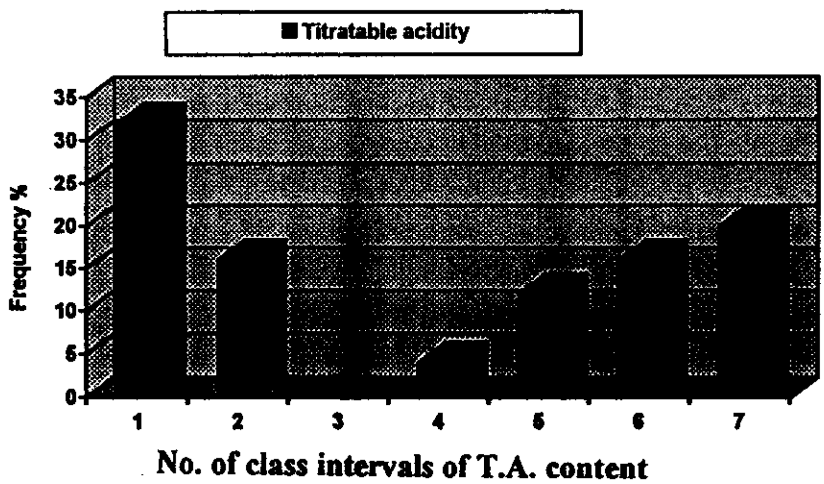

Fig. (2): Frequency distribution of titratable acidity of frozen yoghurts collected from some local markets.

Table (5): Frequency distribution of pH of frozen yoghurts collected from some local markets .

No.	Class intervals	Frequency	
		No. of samples	%
1	5.85-6.06	7	28
2	6.06-6.28	1	4
3	6.29-6.50	1	4
4	6.51-6.72	-	-
5	6.73-6.94	-	-
6	6.95-7.16	4	16
7	7.17-7.38	2	8
8	7.39-7.60	4	16
9	7.61-7.82	5	20
10	7.83-8.04	1	4

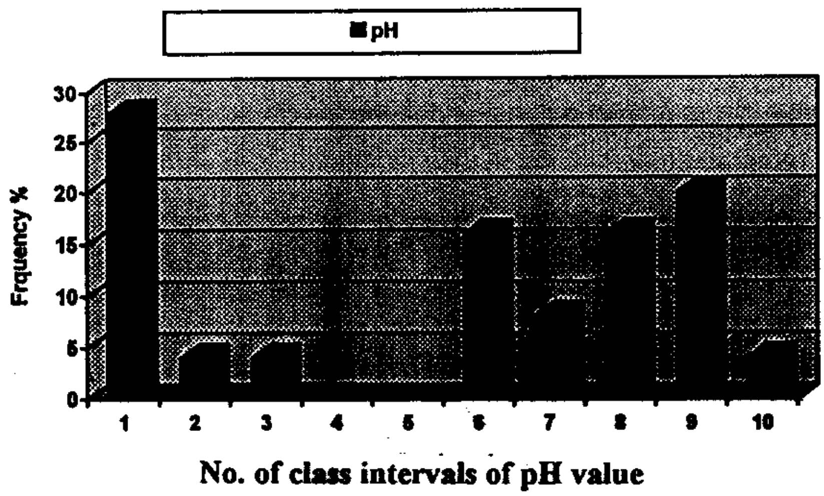

Fig. (3): Frequency distribution on pH of frozen yoghurts collected from some local markets.

Concerning the frequency distribution of pH values in frozen yoghurts examined, Table (5) and Fig. (3) show that, the majority of samples (68%) had a pH level of 6.4 to 7.88. These findings are in agreement with those of Olsen (1990) who mentioned that, frozen yoghurt on the USA markets has a pH level of 6.4 to 7.1, which corresponding to the characteristic pH level of standard milk ice and ice cream.

II- Proximate composition:

1- Fat content:

Results given in Tables (3 and 6) and Fig. (4) show that, there were slightly variations in the fat content between different samples. The fat content of 25 commercial frozen yoghurts represented by 5 Brands ranged from 3.76 to 5.35% with an average of 4.57%. These values were found to lie in the expected range (min. 3.25%) for frozen yoghurt, which represent the generally accepted industry standards in USA (Arbuckle, 1986).

These results are partially in agreement with Kleyn and Elias (1992), and in agreement with those obtained by Tieszen and Baer (1989) who found that, values for fat content in the frozen yoghurt ranged from 1.69 to 5.94%, while higher than those reported by Kosikowski (1981) (0.8 to 2.5%).

The higher frequency (Table 6 and Fig. 4) for samples being 23 out of 25 (92%) lies in the class range of 4.11 to 5.54.

2- Total solids content:

Table (3) shows that although there were slightly wide variations in total solids content between the 25 commercial frozen yoghurts, total solids content fall within the expected range (min. 30%) which represent the generally accepted industry standards in USA (Arbuckle, 1986). The total solids content of 25 frozen yoghurts examined ranged from 31.32 to 38.7% with an average of 35.74%.

The variations of total solids content in frozen yoghurts examined might be attributed to that, most manufacturers resort to use the ingredient available as demanded by consumer buying habits, competitions, costs and finished products quality expected. Generally, the total carbohydrate content might be considered major contributer to the high total solids content in frozen yoghurts examined.

Table (6): Frequency distribution of fat content of frozen yoghurts collected from some local markets .

No.	Class intervals	Frequency	
		No. of samples	%
1	3.76-3.98	2	8
2	3.99-4.10	-	-
3	4.11-4.32	4	16
4	4.33-4.54	3	12
5	4.55-4.76	8	32
6	4.77-4.98	6	24
7	4.99-5.10	-	-
8	5.11-5.32	1	4
9	5.33-5.54	1	4

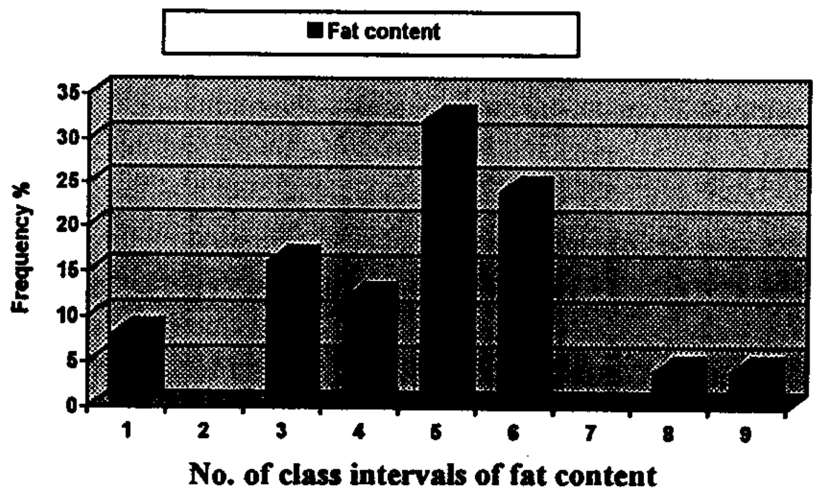

Fig. (4): Frequency distribution of fat content of frozen yoghurts collected from some local markets.

Table (7) and Fig. (5) represent the frequency distribution of total solids content in frozen yoghurts examined. It can be observed that, the highest frequency for frozen yoghurts being 23 out of 25 (92%) lies in the class range of 31.32 to 36.51.

These results are partially in agreement with those obtained by Kosikowski (1981) who found an average total solids content of 30.9% (23.6 to 38.9%), Tieszen and Baer (1989) who found that, values for total solids of commercial frozen yoghurts ranged from 28.83 to 37.6%.

3- Protein content:

The milk proteins contained in ice cream especially in frozen yoghurt are of excellent biological value, because they contain all the essential amino acids. Proteins are essential in human life as components of protoplasm of each living cell. Milk proteins are not only known to be complete but the assimilation of ingested milk proteins is 5-6% more complete than for other proteins in general.

As observed from Table (3) there were slightly variations in protein content between 25 commercial frozen yoghurts examined, where it ranged from 3.53 to 4.52% (averaged 3.89%). These variations in total protein content might be attributed to that, basic formulas of these products might have variations and modifications as demanded by the ingredient available, consumer buying habits, competitions, costs, and finished product quality expected.

The frequency distribution of protein content of frozen yoghurts tested as shown in Table (8) and Fig. (6) show that the majority of frozen yoghurts (96%) lies in the class intervals from 3.53 to 4.40, while 1 out of 25 samples (4%) fall within the class range of 4.52 to 4.62.

The results are partially in agreement with those obtained by Kosikowski (1981) who found that, the protein content of 24 commercial frozen yoghurts ranged from 1.7 to 4.5% and averaged 4%, Tieszen and Baer (1989) who found that, total protein of frozen yoghurt ranged from 1.61 to 4.19%.

4- Ash content:

Table (3) reveals a wide variations in ash content between different samples, where it ranged from 0.69-1.04% with an average of 0.83%.

Invariably, the different Brands of 25 frozen yoghurts established a characteristic ash content range partially independent of total solids or

Table (7): Frequency distribution of total solid contents of frozen yoghurts collected from some local markets .

No.	Class intervals	Frequency	
		No. of samples	%
1	31.32-32.35	2	8
2	32.36-33.39	3	12
3	33.40-34.43	7	28
4	34.44-35.47	10	40
5	35.48-36.51	1	4
6	36.52-37.55	-	-
7	37.56-38.59	1	4
8	38.60-39.63	1	4

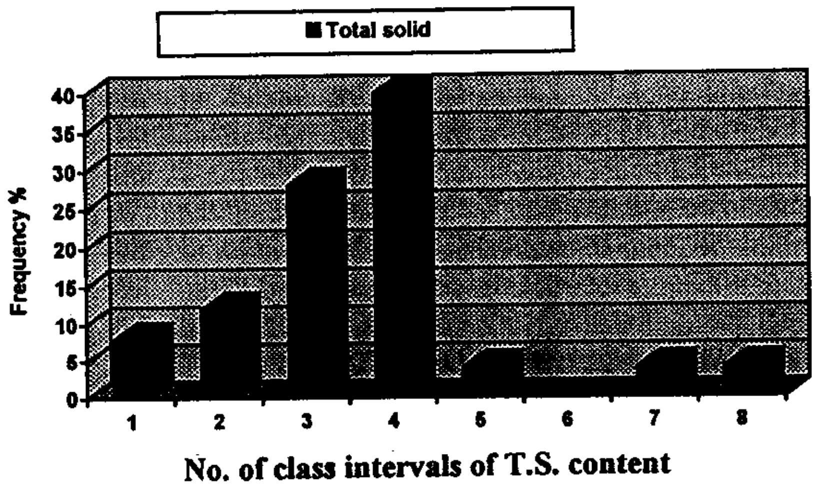

Fig. (5): Frequency distribution of total solid contents of frozen yoghurts collected from some local markets.

Table (8): Frequency distribution of protein content of frozen yoghurts collected from some local markets .

No.	Class intervals	Frequency	
		No. of samples	%
1	3.53-3.63	3	12
2	3.64-3.74	4	16
3	3.75-3.85	4	16
4	3.86-3.96	8	32
5	3.97-4.07	2	8
6	4.08-4.18	1	4
7	4.19-4.29	1	4
8	4.30-4.40	1	4
9	4.41-4.51	-	-
10	4.52-4.62	1	4

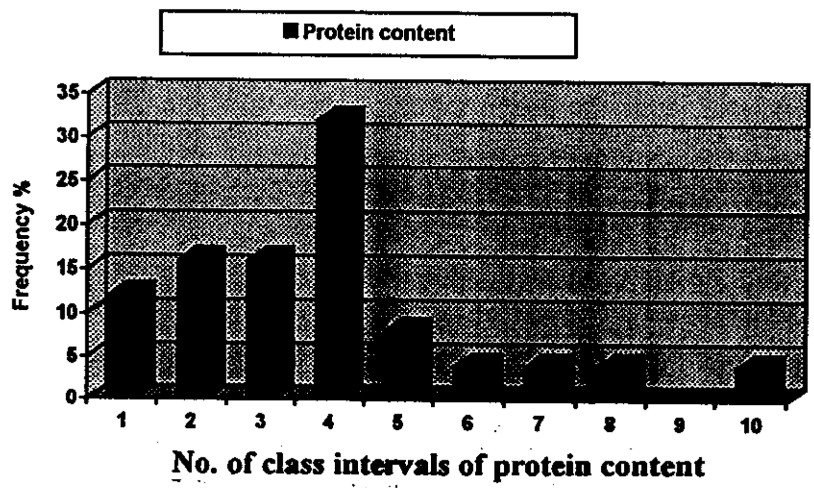

Fig. (6): Frequency distribution of protein contents of frozen yoghurts collected from some local markets.

Table (9): Frequency distribution of ash contents of frozen yoghurts collected from some local markets .

No.	Class intervals	Frequency	
		No. of samples	%
1	0.69-0.74	6	24
2	0.75-0.80	5	20
3	0.81-0.86	6	24
4	0.87-0.92	4	16
5	0.93-0.98	2	8
6	0.99-1.04	2	8

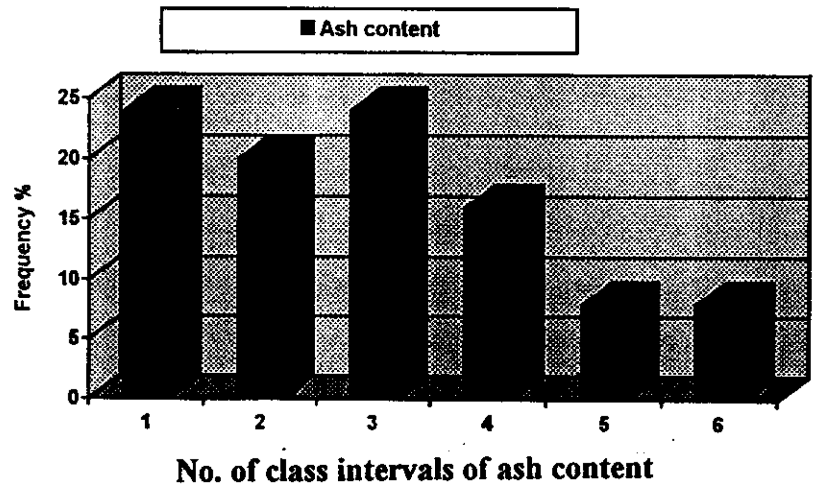

Fig. (7): Frequency distribution of ash contents of frozen yoghurts collected from some local markets.

flavour employed. For examples, sample No. 1 (in Brand vanilla frozen yoghurt) had an ash content of 0.71% and total solids of 34.25, while sample No. 8 had an ash content of 1.04% and total solids of 33.4%; samples No. 12 and 13 had an ash content of 0.74 and 0.81%, and mainly had the same total solids namely 34.85 and 34.60%, respectively; samples No. 14 and 17 contain ash content of 0.83 and 1.01% with total solids of 35.27 and 32.44% respectively; and sample No. 18 and 21 had an ash content of 0.82 and 0.71% with the total solids of 32.35 and 34.6%, respectively.

The higher total solids and ash content indicating the use of basic formulas of high solids not fat. On the other hand, some samples had high ash content with low total solids content, indicating the use of increased amounts of stabilizer/emulsifier mixture, which contains mineral salts and /or bases and salts used as pH controlling agents as previously mentioned under item I-titratable acidity and pH values.

Table (9) and Fig. (7) represent the frequency distribution of ash content in frozen yoghurts examined. It can be observed that, ash content of 25 samples tested fall within the expected ash content of the commercial ice cream and ice milk, which ranged from 0.7 to 1.2% (Arbuckle, 1981).

These results are in agreement with those obtained by Kosikowski (1981) who found an average ash content of 0.9% (0.7-1.2%) in commercial frozen yoghurts; and Tieszen and Baer (1989) who found that, ash content of 19 samples of commercial frozen yoghurts ranged from 0.7 to 1.06%.

5- Total carbohydrates and reducing sugars:

Carbohydrates serves as a source of heat and energy in the body. An overall look to the total carbohydrate in Tables (3 and 10) and Fig. (8) clearly show that, there were a wide variations in the total carbohydrate between different samples, and it was ranged from 19.2 to 26.5%. These values lie in the expected range of total carbohydrate (20-33%) for ice cream, ice milk, sherbets, water ice and frozen yoghurts (Kosikowski, 1981 and Arbuckle, 1986), which represent the generally accepted industry standards in USA. These wide variations in total carbohydrate might be attributed to the variations and/or modifications in basic formulas of these samples as demanded by consumer buying habits, competition, costs and finished product quality expected.

These results are in agreement with the total carbohydrate range previously found by Kosikowski (1981) and Arbuckle (1986).

Concerning reducing sugars, those contain a free potential aldehyde group (aldoses; glucose, galactose and lactose), or contain a free potential ketone groups (ketoses; fructose). Sucrose contains no potentially free aldehyde or ketone group (non-reducing sugar). sucrose (the commonly used sugar in all varieties of ice cream) and lactose (milk sugar; that constitutes over one-third of solid matter in milk and approximatly 20% of the carbohydrate in ice cream) are broken-down to simple sugars and an acid conditions in frozen yoghurt, under the action of lactic acid bacteria or other harmless microorganisms, consequently reducing sugars increased in the product.

As can be seen from Tables (3 and 11) and Fig. (9) and as was the case with the total carbohydrate, there were a wide variations in reducing sugars content between different samples. Reducing sugars content ranged from 3.86 to 9.15% in 25 frozen yoghurt samples examined.

The wide variations in reducing sugars might be attributed to amount and kind of sugars and stabilizers; and amount, type and the activity of the starter cultures used in the manufacture of frozen yoghurts examined.

6- Acetaldehyde content:

The main flavour compounds found in yoghurt, consequently frozen yoghurt are: acetaldehyde, diacetyl, acetone, acetoin and butane-2-one. Acetaldehyde is the main compound recognized as responsible for the flavour intensity in the products (Marshall, 1982).

Data in Table (3) clearly show that, there were a wide variations in acetaldehyde between different Brands, and that acetaldehyde ranged from 2 to 33 ppm. It was higher in Brand Mango frozen yoghurt (33 ppm) and followed by strawberry (22 ppm); chocolate (18 ppm); vanilla from No. 5 to 9 (12 ppm); and the remainder vanilla products and coffee frozen yoghurt (2 ppm).

The wide variations in acetaldehyde between samples might be attributed to the different flavours employed and to using different strains of starter cultures. This observation has been confirmed by Pette and Lolkema (1950) and Tamime and Robinson (1985). However, they observed that the level of acetaldehyde was much greater in mixed culture due to the associative growth of lactic acid bacteria used. They

Table (10): Frequency distribution of total carbohydrates content of frozen yoghurts collected from some local markets .

No.	Class intervals	Frequency	
		No. of samples	%
1	19.20-20.24	1	4
2	20.25-21.29	2	8
3	21.30-22.34	9	36
4	22.35-23.39	9	36
5	23.40-24.44	3	12
6	24.45-25.49	-	-
7	25.50-26.54	1	4

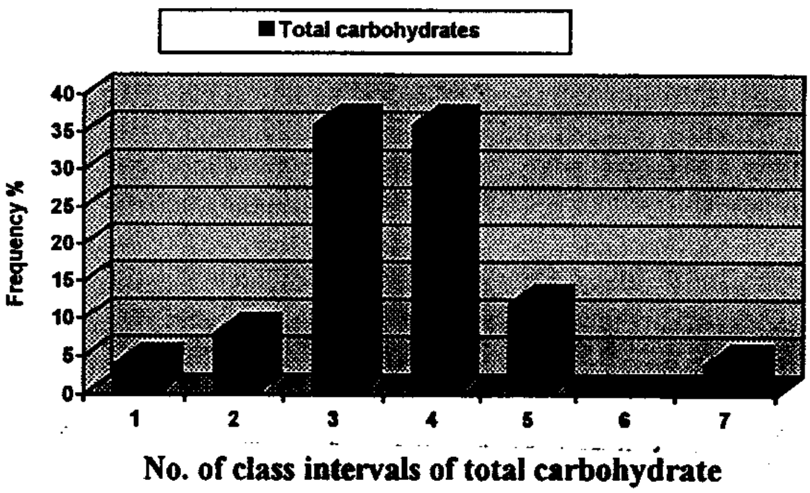

Fig. (8): Frequency distribution of total carbohydrate contents of frozen yoghurts collected from some local markets.

Table (11): Frequency distribution of reducing sugars of frozen yoghurts collected from some local markets .

No.	Class intervals	Frequency	
		No. of samples	%
1	3.86-4.50	3	12
2	4.51-5.15	-	-
3	5.16-5.80	1	4
4	5.81-6.45	3	12
5	6.46-7.10	2	8
6	7.11-7.75	7	28
7	7.76-8.40	4	16
8	8.41-9.05	4	16
9	9.06-9.70	1	4

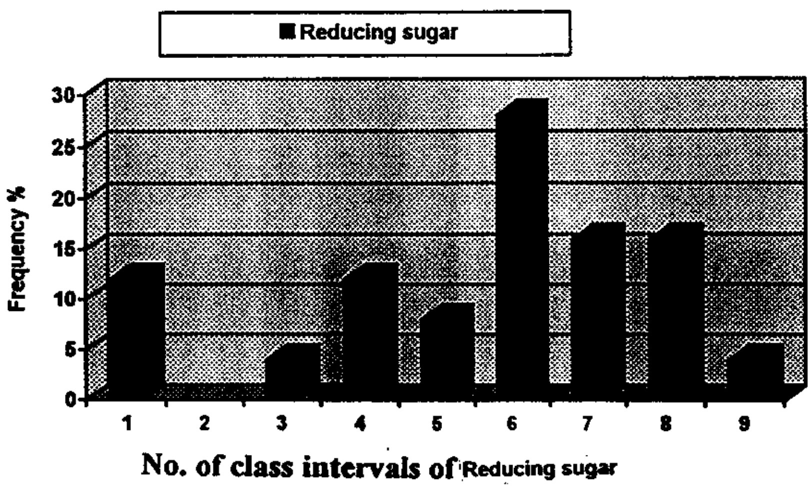

Fig. (9): Frequency distribution of reducing sugar of frozen yoghurts collected from some local markets.

found that, St. salivarius ssp. thermophilus and Lb. delbrueckii ssp. bulgaricus produce acetaldehyde ranged from 1 to 8.3 ppm; and 1.4 to 12.2 ppm, respectively. While mixed cultures produce acetaldehyde ranged from 2 to 41 ppm.

The higher levels of acetaldehyde in some Brands might be attributed to the presence of some milk ingredients required by the starter cultures for producing acetaldehyde such as lactose in higher amounts (Lees and Jago, 1966 and Kennan and Bills, 1968); amino acids threonine and methionine (Wilkins *et al.*, 1986); and from nucleic acids (Lees and Jago, 1978b).

III- Melting resistance:

Regarding melting resistance table (12) shows that, while there were a wide variations in melting resistance, determined as weight loss% at 30°C, between frozen yoghurts examined after 1 and 1½ h., all frozen yoghurt samples have melted after 2h. It was ranged from 41.77 to 59.90% and 78.02 to 94.88% after 1 and 1½ h, respectively. These wide variations might be attributed to the different overrun between these samples.

Invariably, the different Brands (vanilla, strawberry, chocolate, coffee and mango frozen yoghurts) established a characteristic melting resistance range, expressed as weight loss% independent of the flavour employed. For example, strawberry and mango fruits are expected to increase the melting resistance (decrease the weight loss%) of strawberry and mango frozen yoghurts examined than vanilla, chocolate and coffee, due to the combined effect of the lower pH values and highly pectin content of strawberry and mango, which consequently decrease amount of the incorporated air in frozen yoghurt. But the higher weight loss% was found with mango frozen yoghurts (ranged from 88.42 to 94.88% and averaged 92.22%) and strawberry frozen yoghurt (ranged from 88.11 to 91.90% and averaged 89.80%). While, the lower weight loss % was found with coffee frozen yoghurts (ranged 80.47 to 83.53% and averaged 81.44%); chocolate samples (ranged 78.03 to 85.42% and averaged 82.19%). and vanilla frozen yoghurts (ranged 78.02 to 100% and averaged 86.25%).

Table (12): Melting resistance (expressed as weight loss% at 30°C) of frozen yoghurts collected from some local markets.

Sample	Brands	Melting resistance as weight loss% at 30°C		
		1 h	1½ h	2 h
1		48.93	89.25	Complete melting
2		48.84	83.28	"
3		48.49	81.67	"
4		48.61	83.09	"
5	Vanilla	44.62	86.16	"
6		42.60	78.02	"
7		45.60	88.10	"
8		44.80	86.70	"
9		47.00	100.00	"
	Average	46.61	86.25	
10		54.60	91.18	"
11	Strawberry	51.06	88.11	"
12		47.70	91.90	"
13		52.94	88.80	"
	Average	51.58	89.80	
14		44.26	84.41	"
15	Chocolate	46.45	80.89	"
16		46.10	78.03	"
17		43.28	85.42	"
	Average	45.02	82.19	
18		52.03	80.76	"
19	Coffee	51.06	80.47	"
20		59.90	80.98	"
21		55.62	83.53	"
	Average	54.65	81.44	
22		41.77	94.88	"
23	Mango	47.17	93.25	"
24		45.95	88.42	"
25		53.64	92.34	"
	Average	47.13	92.22	

Table (13): Frequency distribution of melting resistance (as weight of loss% at 30°C) of frozen yoghurts collected from some local markets after 1 h.

No.	Class intervals	Frequency	
		No. of samples	%
1	41.77-44.26	4	16
2	44.27-46.76	6	24
3	46.77-49.26	7	28
4	49.27-51.76	2	8
5	51.77-54.26	3	12
6	54.27-56.76	2	8
7	56.77-59.26	-	-
8	59.27-61.76	1	4

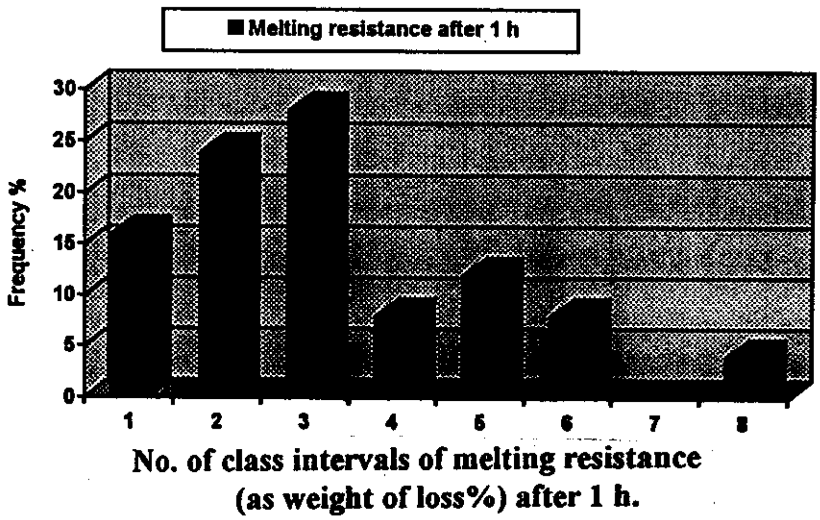

Fig. (10): Frequency distribution of melting resistance (as weight of loss % at 30°C) of frozen yoghurts collected from some local markets after 1h.

Table (14): Frequency distribution of melting resistance (as weight of loss% at 30°C) of frozen yoghurts collected from some local markets after 1½ h.

No.	Class intervals	Frequency	
		No. of samples	%
1	78.02-80.47	3	12
2	80.48-82.93	4	16
3	82.94-85.39	4	16
4	85.40-87.85	3	12
5	87.86-90.31	5	20
6	90.32-92.77	3	12
7	92.78-95.23	2	8
8	95.24-94.69	-	-
9	94.70-97.15	-	-
10	97.16-99.61	-	-
11	99.62-102.07	1	4

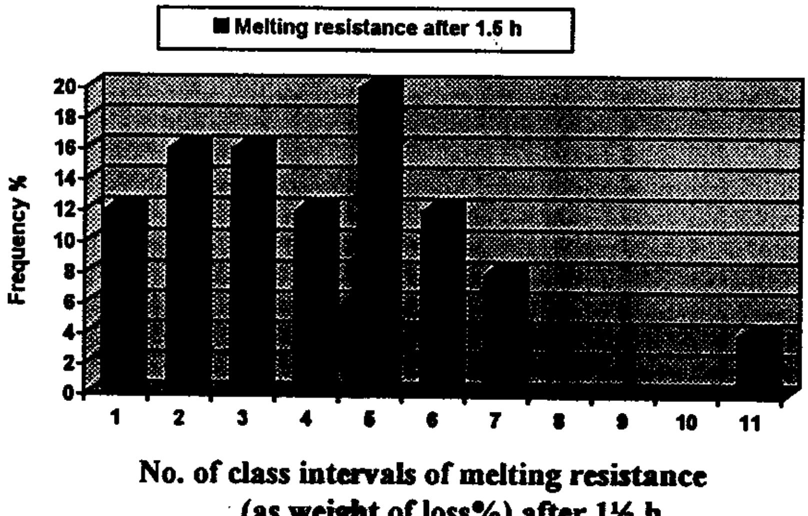

Fig. (11): Frequency distribution of melting resistance (as weight of loss % at 30°C) of frozen yoghurts collected from some local markets after 1½h.

As might be expected from these findings that, strawberry and mango used in frozen yoghurt examined were not natural flavouring fruits, but might be artificial flavouring substances.

The higher frequency distribution of melting resistance as weight loss% at 30°C (Tables, 13 and 14 and Figs., 10 and 11) of frozen yoghurts examined being 24 out of 25 samples (96%) lies in the class range of 41.77 to 56.76 and 78.02 to 95.23 after 1 and 1½ h, respectively, while being 1 out of 25 samples (4%) has complete melted after 1½ h.

V- Microbiological properties:

a- Lactic acid bacteria:

(*St. salivarius* ssp. *thermophilus* and *Lb. delbrueckii* ssp. *bulgaricus*).

In recent years there has been a noticeable consumption of frozen yoghurt in local Egyptian market. However, there are no specification for the product in regard to type of strains used. In the present part attempts have been made to determined lactic acid bacteria used in commercial frozen yoghurt manufacture.

as shown in Table (15) total counts of *St. salivarius* ssp. *thermophilus* of 25 frozen yoghurts examined ranged from $13x10^5$ to $103x10^5$ with an average of $41.84x10^5$. Corresponding counts for *Lb. delbrueckii* ssp. *bulgaricus* were ranged from $15x10^5$ to $88x10^5$ with an average of $46.2x10^5$. These results also indicate that, the desired ratio of 1 : 1 between the two organisms existed only in one sample, No. 14 (4%), while all other samples had a different ratios. Of the 25 frozen yoghurts examined, only 7 samples (28%) had a ratio of more than 1 : 1 in favour of *St. salivarius* ssp. *thermophilus*. The fact that *St. salivarius* ssp. *thermophilus* responded more vigorously in mixed culture was attributed to the proteolytic activity of *Lb. delbrueckii* ssp. *bulgaricus* notably through the release of valine, histidine, histidine plus glycine, leucine, isoleucine, methionine, phenylalanine, arginine, tryptophane and glutamic acid - valine most active (Robinson, 1990).

On the other hand, 17 samples (68%) were in favour of *Lb. delbrueckii* ssp. *bulgaricus*. This obviously explain the stimulation of *Lb. delbrueckii* ssp. *bulgaricus*, which attributed to a factor originating from the metabolic activity of *St. salivarius* ssp. *thermophilus* "formic acid and carbon dioxide" (Robinson, 1990).

These findings are partially in line with those obtained by Kosikowski (1981) who found that, bacterial cells appeared usually equally divided in numbers between cocci and rods or possessing about 70% cocci and 30% rods.

Table (16) and Fig. (12) show the frequency distribution of the counts of both *St. salivarius* ssp. *thermophilus* and ***Lb. delbrueckii*** **ssp.** ***bulgaricus*** in 25 frozen yoghurts represented by 5 Brands. It appears that, the majority of samples (76%) had a count of *St. salivarius* ssp. *thermophilus* between lb to $63x10^5$, and only 3 samples (12%) had counts in the range of 64 to $111x10^5$ for *Lb. delbrueckii* ssp. *bulgaricus*. These counts are partially in agreement with those obtained by Kosikowski (1981) who found that, total microscopic count numbers of 23 frozen yoghurts examined ranged from less than $3.3x10^5$ to $64x10^7$ per gram (as coccus and rod bacteria); Tieszen and Baer (1989) who found that, 14 out of 19 frozen yoghurts were well blew the advised limits of $5x10^6$ to $7x10^6$/g for possible health benefits; Brown *et al.* (1991) who found that, 97 samples of frozen yoghurt (hard and soft), respresenting 24 Brands, showed mean value of $2.5x10^5$ cfu lactic acid bacteria/g (ranged from $4.9x10^6$ in hard frozen yoghurts to 1 x 10^5/g in soft frozen yoghurts); and Whitehead *et al.* (1993) who found that, all (20 frozen yoghurts) but 3 samples were found to contain lactic acid bacteria (yoghurt culture) in concentration >$1x10^6$ cfu/g.

b- Coliforms and *Escherichia coli* count:

The coliforms and presumptive *E. coli* test still remains an extremely useful laboratory test for monitoring ice cream hygiene for public health purposes. The results are satisfactory if presumptive coliform absent from 0.01 g in Scotland and UK standards (Robinson, 1990); while the IDF and Egyptian standards requirements of less than 100 cfu/g (Robinson, 1990) and 10 cfu/g of coliforms, respectively, with *E. coli* absent from 1 gram. The International Commission on Microbiological Specifications for Foods, ICMSF (1974) suggests the following coliform limits for ice cream: (1) values equal to 10 cfu/g or less are acceptable, above is defective or marginally acceptable, but a count above 1000 cfu/g is not accepted; (2) in ice cream with added ingredients (complex), values equal to or less 100 cfu/g are acceptable, above is defective or marginally acceptable, but a count above 1000 cfu/g

Table (15): The lactic acid bacterial counts of frozen yoghurts collected from some local markets.

Sample No.	Type	*St. salivarius* ssp. *thermophilus*	*Lb. delbrueckii* ssp. *bulgaricus*	Ratio St : Lb
		No. x10^5 cfu/ml		
1	Vanilla	46	50	1 : 1.1
2		52	76	1 : 1.5
3		57	62	1 : 1.1
4		26	54	1 : 2.1
5		20	54	1 : 2.7
6		13	30	1 : 2.3
7		35	29	1.2 : 1
8		19	28	1 : 2.0
9		13	33	1 : 2.5
10	Strawberry	83	88	1 : 1.1
11		57	79	1 : 1.4
12		50	43	1.2 : 1
13		22	63	1 : 2.9
14	Chocolate	47	49	1 : 1.0
15		38	46	1 : 1.2
16		50	53	1 : 1.1
17		27	30	1 : 1.1
18	Coffee	54	45	1.2 : 1
19		14	32	1 : 2.3
20		68	57	1.2 : 1
21		61	46	1.3 : 1
22	Mango	33	23	1.4 : 1
23		42	49	1 : 1.2
24		16	21	1 : 1.3
25		103	15	7 : 1
Average		**41.84**	**46.20**	

Table (16): Frequency distribution of *St. salivarius* ssp. *thermophilus* and *Lb. delbrueckii* ssp. *bulgaricus* counts of frozen yoghurts collected from some local markets.

No.	Class intervals No. x10^5 cfu/ml	*St. salivarius* ssp. *thermophilus* Frequency		*Lb. delbrueckii* ssp. *bulgaricus* Frequency	
		No. of samples	%	No. of samples	%
1	0-15	3	12	1	4
2	16-31	6	24	6	24
3	32-47	6	24	6	24
4	48-63	7	28	9	36
5	64-79	1	4	2	8
6	80-95	1	4	1	4
7	96-111	1	4	-	-

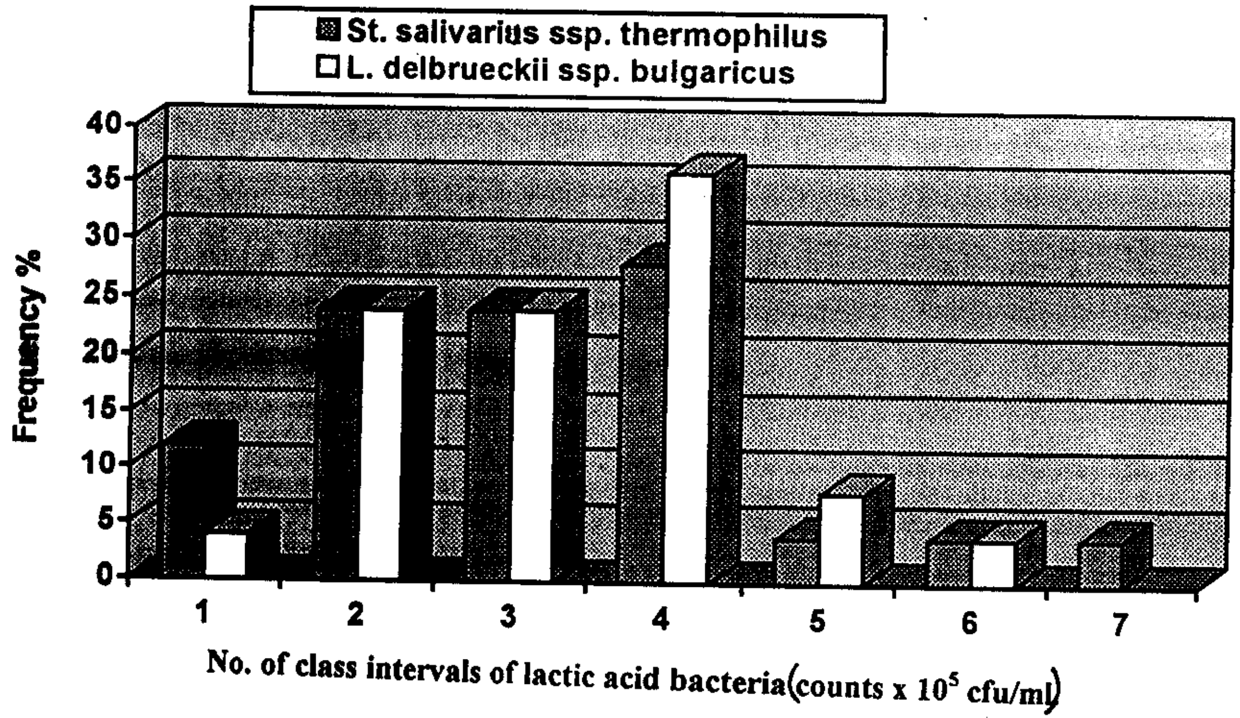

Fig. (12): Frequency distribution of *S. salivorias* spp. *thermophilus* and *Lb. delbrueckii* ssp. *bulgaricus* counts of frozen yoghurts samples collected from some local.

Table (17): The total coliforms, E. coli and yeasts & moulds counts per ml of frozen yoghurts collected from some local markets .

Sample No.	Type	Coliform	E. coli	Yeasts & moulds
		cfu/ml		
1	Vanilla	30	3	20
2		12	-	60
3		20	-	20
4		20	-	10
5		15	2	50
6		11	-	34
7		20	-	32
8		20	-	60
9		20	-	30
10	Strawberry	20	4	40
11		30	-	40
12		21	-	40
13		23	-	63
14	Chocolate	20	5	40
15		13	-	30
16		20	-	40
17		13	-	50
18	Coffee	20	1	30
19		13	-	40
20		20	-	50
21		12	-	40
22	Mango	20	-	40
23		24	3	30
24		20	-	40
25		20	-	40
Average		**19.1**	-	**38.76**

is not accepted. It is felt that these values are standards which could be applied in many, if not all, countries.

It might be observed from Table (17) that the coliforms were detected in all frozen yoghurts examined and exceeded the Egyptian coliform standards of less than 10 cfu/g, with *E. coli* absent from 1 g. The coliforms ranged from 11 to 30 cfu/ml with an average of 19.1 cfu/ml. The lower counts is most probably due to the higher levels of T.A. and to the higher counts of *Lb. delbrueckii* ssp. *bulgaricus*, which is reported to have a suppressing effect on the coliforms (Harby, 1985). In a study to evaluate the microbial quality of frozen yoghurts, Brown *et al.*, (1991) found that during summer frozen yoghurts exceeded the coliform standard in 24% of the 76 samples tested.

Concerning *E. coli*, results given in Table (17) show that, although 25 frozen yoghurt samples tested contained coliform organisms, only 6 samples (24%) contained *E. coli*. The high counts of coliforms and presence of *E. coli* in some frozen yoghurts indicate that, the production of frozen yoghurt occurs under unfavorable conditions and that, the heat treatment undertaken might be insufficient and/or recontamination occurs after the heat treatment, but the final selling point may also be involved.

These findings indicate that, some concern is warranted over the microbial quality of frozen yoghurt as offered for sale.

Data presented in Table (18) and Fig. (13) show the frequency distribution of coliform counts in 25 frozen yoghurts examined. It indicates that, the majority of samples (84%) had a count of not more than 22 cfu/ml, while 4 out of 25 samples (16%) had a count ranged from 23 to 30 cfu/ml.

c- Yeasts and moulds count (Y & M):

Yeasts and moulds in particular, are little affected by low pH, and with both sucrose and lactose available as energy sources, spoilage can occur rapidly. Yeasts, in particular, are a major concen, for while the lactose-utilizing yeasts (Kluy veromyces makxianus var. lactis) can build up on plant surfaces. Contamination of the more popular types of stirred yoghurt and frozen yoghurt can be caused by the fruits and nuts. However, Barnes *et al.* (1979) have proposed the following:

Target figure	< 10 cfu (Y & M/g)
Acceptable	10-50 cfu (Y & M/g)
Acceptable (but to be improved upon)	50-100 cfu (Y & M/g)

Unacceptable 100 cfu (Y & M/g)

However, Robinson (1990) have suggested > 10 Y & M cell/g in ice cream (as target figure).

As shown in Table (17) the yeasts and moulds were detected in all frozen yoghurts examined. It was ranged from 10 to 63 cfu/ml with an average of 38.76 cfu/ml. These values were found to lie in the acceptable range (10-50 cfu/ml) and acceptable range, but to be improved upon (50-100 cfu/ml) suggested by Barnes et al. (1979).

These findings again indicate that, the production of frozen yoghurts examined might be occurs under unfavorable conditions and that the heat treatment under-taken might be insufficient and/or recontamination occurs after the heat treatments. Contamination can also be caused by the fruits, nuts and sucrose. The final selling point may also be involved.

The frequency distribution of the counts of Y & M as shown in Table (19) and Fig. (14) shows that the majority of the frozen yoghurts examined (64%) lies in the class intervals from 21 to 42, while, 4% and 12% of the samples lie in the class intervals from 10 to 20 and from 43 to 64, respectively.

From this part it can be concluded that commercial frozen yoghurt, abnormally high in pH which possess low lactic acid bacteria, if incubated, may permit contaminating bacteria to grow to large numbers and pose potential health hazard, as for example, all frozen yoghurts tested had high counts of coliforms and presence of *E. coli* in some frozen yoghurts (24%). Therefore, there is no value which may be easily attained in the frozen yoghurt without developing enough lactic acid. Therefore, these results have shown the need to use an other effective species and strains produce metabolites, to produce frozen yoghurt of antibiotic role, beneficial therapeutic effect and increased shelf-life, even with products of high pH and low titratable acidity.

Table (18): Frequency distribution of coliform counts of frozen yoghurts collected from some local markets .

No.	Class intervals	Frequency	
		No. of samples	%
1	11-14	6	24
2	15-18	1	4
3	19-22	14	56
4	23-26	2	8
5	27-30	2	8

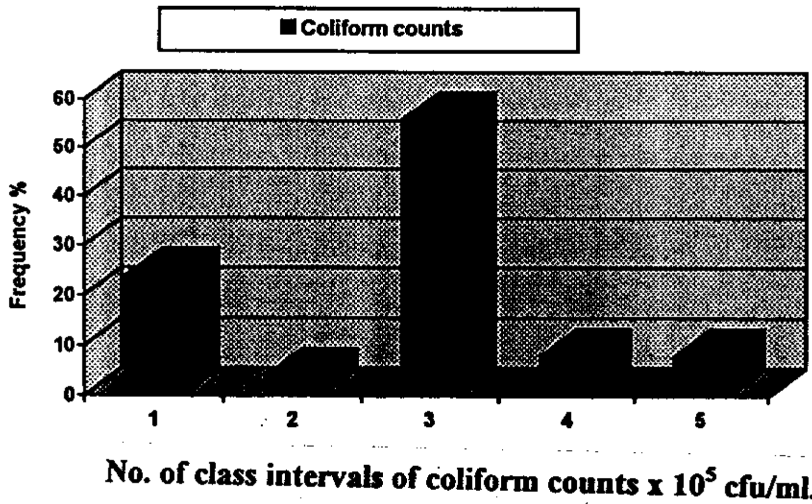

Fig. (13): Frequency distribution of coliform counts of frozen yoghurts collected from some local markets.

Table (19): Frequency distribution of yeasts and moulds of frozen yoghurts collected from some local markets .

No.	Class intervals	Frequency	
		No. of samples	%
1	10-20	3	12
2	21-31	4	16
3	32-42	12	48
4	43-53	3	12
5	54-64	3	12

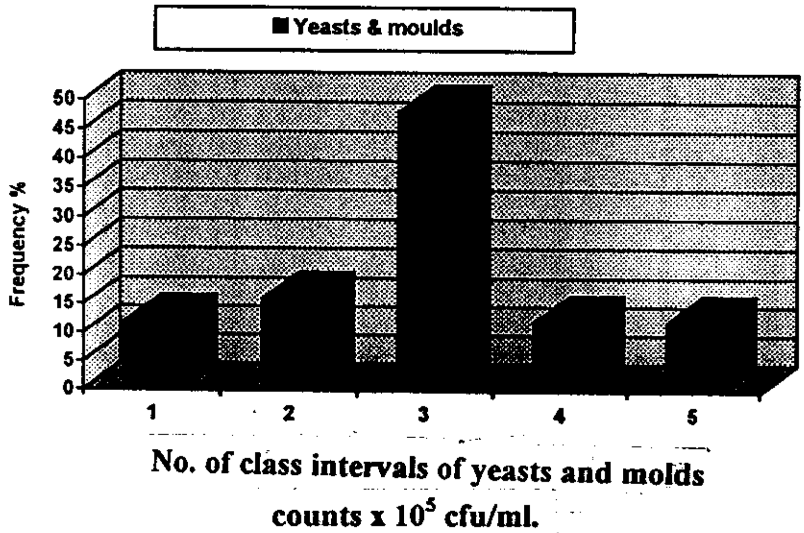

Fig. (14): Frequency distribution of yeasts and moulds of frozen yoghurts collected from some local markets.