

EVALUATION OF THE MICROBIOLOGICAL QUALITY OF MILK AND YOGHURT BLENDED WITH SOME FRUITS JUICE

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ABSTRACT

A study was describes that plain yoghurt were prepared in the laboratory scale production from cow's milk obtained from certified dairy farm in Cairo – Egypt .Plain yoghurt was blended with fresh juice (Guava juice- Mango juice- Strawberry juice) and commercial yoghurt fortified by juice. The chemical and microbiological quality of yoghurt, yoghurt juice blends and fresh juice samples were investigated during refrigerated storage at 4 0 C for 2 weeks, and 6 months for juice. The microbial analyses influence the growth of Yeast & mould, coliform organisms while lactobacillus bulgaricus and streptococcus thermophilus were not significantly affected before and after storage. The result of the study showed that fresh juice had significant effect on acceptability of yoghurt before and after storage.

INTRODUCTION

Yoghurt is a one of the best-known of the food that contain probiotics. Which is defined by the codex Alimentarius of 2003 as a coagulated milk product that results from the fermentation of lactic acid in milk by *Lactobacillus delbrueckii* ssp. *Bulgaricus* and *streptococcus thermophilus*? That the nutrient composition of yoghurt is based on the nutrient composition of milk. So yoghurt is a very popular fermented milk product that is basically produced by the fermentation of pasteurized (full or skimmed) milk, widely consumed all over the world. To preserve its inherent quality and sensory characteristics, blending with juice is essential. The influence of blending fruit juices (Guava, Mango, and Strawberry) on the sensory and physicochemical characteristics of yoghurt. Yoghurt is a highly nutritious protein-rich product obtained by fermentation of milk with *S. thermophilus* and *L. bulgaricus*. The product is highly acceptable to consumers because of its flavour and aroma, mainly attributed to acetaldehyde, and its texture. The shelf life of yoghurt is short, i.e., 1 day under ambient condition (25–30 °C) and around 5 days at 7°C (Salji *et al.*, 1987), which hinders its commercialization. Yoghurt is maintained at 2–4°C throughout the distribution chain, which not only avoids risk of spoilage from yeasts and moulds but also prevents further activity by starter culture. This, however, adds to the cost of the product. In addition to its high nutritional value, yoghurt possesses antagonistic and therapeutic values. The valuable sensory characteristics of yoghurt are due to its content of carbonyls, mainly acetaldehyde, acetone, acetoin, diacetyl and ethanol,

produced by yoghurt bacteria (Gilliland, 1991). Yoghurt provides higher levels of protein, carbohydrate; calcium and certain B vitamins than milk (Gurr, 1987; Deeth and Tamime, 1981). Several health benefits have been claimed to be associated with the consumption of fermented milk products (Le *et al.*, 1986; Van? Veer *et al.*, 1989; Modler, 1990; Hughes and Hoover, 1991; Kanbe, 1992; Mital and Garg, 1992; Nakazava and Hosono, 1992; Yamamoto *et al.*, 1994). Although yoghurt microflora (*Streptococcus thermophilus* and *Lactobacillus delbrueckii* ssp. *bulgaricus*) have been found to be beneficial for human health and nutrition (Deeth and Tamime, 1981; IDF, 1984). Lactic acid bacteria have been paid increasing attention because of their beneficial effects for the health of their host, and are called probiotics (Fuller, 1989; Prasad, Gill, Smart, & Gopal, 1998). In order to act as probiotics, the bacteria should be delivered alive to the intestine of their host. However, the bacteria are damaged by the digestive juices, such as the acidic stomach solution and bile during the delivery (Kimoto, Ohmomo, Nomura, Kobayashi, & Okamoto, 2000; Vinderola & Reinheimer, 2003). Some methods have been proposed to protect them from the juices; coating the bacteria with oils, proteins or polysaccharides, and inclusion in gels (Lian, Hsiao, & Chou, 2003; Picot & Lacroix, (Matsuo *et al.*, 2009). Fruit juices are becoming an important part modern diet in many communities. In a recent study on food sources of nutrients in the diet of Spanish children fruit juices and citrus fruit were shown to be the principal sources of vitamin C accounting for 43 % (Royo-Bordonada *et al.*, 2003). In another study fruit and juice intake was found to be associated with higher dietary status index in rural women (Knol & Haughton, 1998). In the past it was believed that the acidity of certain foods such fruit juices and yogurt prevent the multiplication and survival of microbes including food borne pathogens and therefore render such foods safe for human consumption. Today this belief is no longer true. Studies have shown that pathogenic bacteria can survive pH as low as 2.5 for 2 h or more (Benjamin & Datta, 1995; De Jonge, Takumi, Ritmeester, & Leusden, 2003; Gordon & Small, 1993; Lin *et al.*, 1996). Furthermore, in the last decade a number of outbreaks of *Escherichia coli* O157:H7, *Salmonella*, *Cryptosporidium* and Norwalk virus infections associated with consumption of unpasteurized fruit juice have been documented in Europe, United States and other countries (Bresser, Lett, & Weber, 1993; CDC, 1997; Centers for Disease Control (CDC), 1999; Cody, Glynn, & Farrar, 1999; Cook, Dobbs, & Hlady, 1998).

Strawberry is one of the world's largest fruit crops, Strawberry is one of the most delicate and highly perishable fruits, it could consume fresh or in many other forms such as juice, concentrate jam, and jelly and fruit juices with yoghurt and bakery products. Guava being a climacteric fruit exhibits a rise in respiration and ethylene production during ripening, and is highly perishable in nature (Singh and Pal, 2008a, b). Mangoes belong to the genus *Mangifera*, The fruit pulp is high in prebiotic dietary fiber, vitamin C, polyphenols and provitamin A carotenoids. Acidified milk drinks (AMDs) are a diverse group of beverages including drinking yoghurts and milk/juice drinks (Nakamura, Yoshida, Maeda, & Corredig, 2006). A common denominator of these products is their low pH and low viscosity, which results in

sedimentation problems due to aggregation of milk protein (Amice-Quemeneur Haluk, Hardy, & Kravtchenko, 1995). Drinking yoghurts are made by diluting a fermented yoghurt base with water (and often fruit concentrate as well) whereas milk–juice drinks are made from diluted fruit concentrate and milk powder. And the remainder milk–juice drinks (made from fruit concentrate and reconstituted milk powder), were submitted to descriptive sensory analysis and rheological characterization. (Janh *et al.*, 2008).

These microorganisms must be viable, active and abundant in the product at the date of minimum durability. If the product has been subjected to heat treatment after the fermentation the requirement for the viability of the micro-organisms is no longer applicable (FAO/OMS, 2003) The objectionable contamination of the examined samples with the different types of coliforms and enterococci, as well as other contaminates is never desirable and may be responsible for loss quality and spoilage of the products that render them at times inedible and these micro-organisms such as coliform, yeast & mould, E.coli .Therefore the present work was designed to study the quality of yoghurt by juice blending to improve micro-organism characteristics and safety of blends of juice yoghurt drinks and compared with commercial juice yoghurt drinks.

The scope of the investigation was concentrated on the following main aspects:-

- * Study the effect of Micro-organism of yoghurt, blends of juice yoghurt drinks, juice.
- * Study the effect of storage time at 4⁰C for 6 months of yoghurt, juice, blends of yoghurt juice drinks.

MATERIALS AND METHODS

Source of fruit samples:

Commercially grown Mature Guava (*Psidium guajava* L.), mango (*Mangifera indica* L.) and Strawberries (*Anna delicious*) were used for this study. These fruits were in the early ripening stage (green yellow color of both orange and mango, but red color of strawberries) and were processed on the same day of purchasing from a local supermarket in Cairo, Egypt or kept at 3-4⁰ C until needed.

Preparation of fruit material:

The selected fruits had a good maturity, color, free from any undesirable odor, free from any spoil part by microorganisms or enzymes or accidents from transporting process and/or premature or have increasing in maturity.

Each orange fruit was rinsed with water, sectioned to half slices. Each strawberry fruit was rinsed with water, sectioned to slices at least 1.0 – 2.0 cm from the skin end (to exclude the effects of bruising), exposing fresh surface. Each mango fruit was simultaneously peeled and sliced to 0.5 -1.0 cm thick slices, then immediately placed in glass beakers.

Extraction of fruit juices:

Fruit juicing was performed at room temperature. Orange, mango and strawberries fruits were sanitized before making juice by immersing for 1min. in 200ppm Cl₂ (Sodium hypochlorite solution, NaClO) and then rinsing with water to remove the Cl₂ residue.

All of the equipment and glassware used to produce the juice were sanitized by immersion in 1000ppm Cl₂ (Sodium hypochlorite solution, NaClO), pH 6.5 (adjusted with citric acid) for 1min and then rinsed with water to remove the residue.

All containers in which the juice was to be held were autoclaved in a AMSCO Scientific, SV-120, (USA) at 121° C for 30min.

1-1- Guava (G) and Mango (M) fruits were rinsed with water, sectioned to longitudinal slices, and juiced with an Acme Supreme Juicerator Model 6001 (Acme Juicer Mfg. Co., Lemoyne, PA) lined with a 46 x 57cm strip of Whitman No.1 filter paper. Juice was collected in a beaker containing 1% antifoam emulsion (Sigma Chemical Co, St Louis, MO), to prevent foaming during extraction of the juice, and ascorbic acid (5mg/100ml juice) with stirring.

1- 2- Strawberries (S) fruits were rinsed with water, cut into small pieces and pureed in a Waring blender for 2-3min., then extracted by cheese cloth and storage at glass for 6 months at 4°C.

Storage fruit pulp freezing:

Each one concentration or time of thermal, chemical and natural extracts pretreatment including banana and apple treated and untreated were blended with stab mixer (Braun Type 4169, Spin) to obtain the required apple or banana pulp and packed in glass bottles and stored at -18°C in frozen storage until sample analysis (PPO, POD and CAT enzyme activities non-enzymatic browning, color, vitamin C, total carotenoids and microbiology) which was carried out at 0, 2, 4, 6 and 8 weeks of frozen storage.

Collection of samples

- a) The study includes examination of sixty six samples of yoghurt representing: (i) 6 samples lab. made plain yoghurt; (ii) 6 random samples of market plain yoghurt; (iii) 18 samples of lab. made fortified yoghurt, representing 6 samples of fruit juices each of Guava, Mango & Strawberry; (iv) 18 samples of market made plain yoghurt fortified in the lab. Using Guava juice, Mango juice & Strawberry juice (6 samples each); (vi) 18 samples of fortified market made yoghurt including Guava, Mango & Strawberry (6 samples each).
- b) 18 samples of lab. made fruit juices representing 6 samples each of Guava, Mango & Strawberry.

Milk used for preparation of lab. made yoghurt:

The raw buffalo's milk used for lab. made yoghurt was obtained from a certified dairy farm at sharkia Governorate to ensure its freedom from inhibitory substance.

Starter cultures used for lab. made plain yoghurt:

Old plain yoghurt obtained from HACCP, certified & ISO22000: 2005 Dairy Company was used as a source of the starter culture.

Preparation of lab. made yoghurt:

Prepare raw buffalo milk from certain supermarket in Cairo, Egypt. was subjected to a heat treatment at 92°C for 20 min to kill microorganisms and to evaporate 25% from water content in milk. And left for cooling to 40 – 45°C. As starter culture yoghurt, 1 day old yoghurt was added to the milk followed by mixing, which was then packed in presterilized glass capped cups 100ml capacity, followed by incubation at 42°C for 3-4 hours till gel forms (pH 4.5). Freshly made yoghurt was cooled and stored at refrigeration temperatures 5°C till examination to slow down the physical, chemical and microbiological degradation.

Preparation of fruit juice:

Guava, Mango, Strawberry fruits were procured from the local fruit market. The fruits were washed, peeled, crushed and passed through pulper to obtain pulp. In case of Guava, Mango, Strawberry were peeled and passed through a screw type juice extractor to obtain Guava, Mango, Strawberry juice stored and refrigeration at 4°C temperature for 6 months and were analyzed for colour, Total Soluble Solid, PH, Titrable Acidity, Viscosity, Total Phenol, vitamin C, at regular intervals 1 month.

Preparation of lab. Fortified yoghurt:

As the same preparation of yoghurt then adding fruit juice so drinking yogurt is essentially stirred yogurt that has a sufficiently low total solids content to achieve a liquid or pourable consistency and which has undergone homogenization to further reduce the viscosity. Fruit and flavour may be incorporated at this time, and then packaged. The product is now cooled and stored at refrigeration temperatures (5°C) to slow down the physical, chemical and microbiological degradation. Sweeteners, flavouring and colouring are invariably added.

Analytical methods

Microbiological Examination of samples

Preparation of samples for examination:

Preparation of yoghurt samples: (APHA, 1992)

The collected yoghurt samples were prepared for micro-biological examination according to American public health Association (APHA, 1992). Preparation of food homogenate and decimal dilutions, (APHA, 1992)

Aseptically 25g from each sample were homogenized in a sterile stomacher bag with 225 ml of Ringers solution. One ml of the previously prepared well-mixed first dilution was transferred to sterile test tube containing 9 ml of sterile diluent and mixed to obtain 1/100 dilution, from which one ml was added to another 9 ml sterilized diluents to obtain further ten fold serial dilutions.

The prepared samples were subjected to the following microbiological examinations

Preparation of fruit juice samples: (APHA, 1992)

The collected of fruit juice samples were prepared for micro-biological examination according to American public health Association (APHA, 1992). Preparation of 10 folds decimal dilution .

Determination of Aerobic Plate Count:

The total counts of the aerobic mesophilic bacteria was determined using the total plate count method, standard plate count agar (Oxoid Ltd, Basingstoke, Hampshire – England). The number of colonies was counted and recorded as colony forming units per gram of sample (cfu /g).

Determination of Yeast and Mould Count: (ISO, 1994)

Duplicate plates of chloramphenicol yeast extract agar were inoculated with 0.1 ml of previously prepared serial dilutions and evenly spread on to the surface of agar plates. Inoculated plates were incubated at 25 °C for 3 to 5 days. The first examination was done after 3 days of incubation to determine the degree of mould growth. After 5 days, yeast as well as mould colonies were enumerated on countable plates separately. The yeast and mould count –g of examined samples was calculated and recorded.

Determination of Escherichia coli content (MPN/g) using E. coli- MUG method (ISO, 1994)

One ml portion from each of the previously prepared decimal dilutions was inoculated into a series of 3 fermentation tubes containing E. coli broth-MUG, supplemented with inverted Durham's tubes for collection of gas. Inoculated tubes as well as control one were incubated at 35°C for 48 ± 2 hours. Gas positive tubes (Coliforms positive) were exposed to long wave (365nm) UV light; positive MUG exhibits a bluish fluorescence that is easily visualized in the medium. Calculate and record the MPN/g of Escherichia coli in the samples examined.

Determination of Coliform Count (MPN/g):

One ml of prepared sample and from each of the previously prepared decimal dilutions was inoculated into a series of 3 fermentation tubes containing Lauryl Sulphate Tryptose broth (LST) supplemented with inverted Durham's tubes for collection of gas. Inoculated tubes as well as control one were incubated at 35°C for 48 ± 2 hours, and then examined for gas production. MPN/g of the samples examined was obtained from the results recorded.

RESULTS

Table (1): Incidence of Coliforms in examined lab made yoghurt samples.

Type of samples	No of Samples	Positive samples	%
Lab made plain yoghurt	6	2	33.33
Lab made yoghurt fortified with Guava	6	4	66.76
Lab made yoghurt fortified with Mango	6	3	50.00
Lab made yoghurt fortified with Strawberry	6	2	33.33

Table (2): Incidence of Coliforms in examined market yoghurt samples

Type of samples	No of Samples	Positive Samples	%
Market plain yoghurt	6	4	66.66.
Market fortified yoghurt by Guava	6	3	50.00
Market fortified yoghurt by Mango	6	5	83.33
Market fortified yoghurt by Strawberry	6	4	66.66

Table (3): Incidence of Coliforms in examined Juice samples.

Type of samples	No. of samples	Positive samples	%
Guava Juice	6	4	66.76
Mango Juice	6	3	50
Strawberry Juice	6	2	33.33

Table (4): Statistical Analytical Results of Microbiological Examination of examined lab made yoghurt Samples based on their Coliform MPN count/gm.

Type of samples	No of Samples	+ve Samples	Min	Max	Mean	S.E.M ±
Lab made plain yoghurt	6	2	10x10	9.8x10 ⁶	6.35x10 ⁵	2.95x10 ⁵
Lab made yoghurt fortified with Guava	6	4	37x10	5.5x10 ⁵	6.35x10 ⁴	2.78x10 ⁴
Lab made yoghurt fortified with Mango	6	3	5.1x10	3x10 ⁷	2.71x10 ⁶	1.32x10 ⁶
Lab made yoghurt fortified with Strawberry	6	2	43x10	2.2x10 ⁷	13.06x10 ⁵	6.90x10 ⁵

Table (5): Frequency distribution of examined Lab made yoghurt samples based on their coliform count/gm.

Intervals	No of positive samples	%
10-10 ²	2	18.18
10 ² -10 ⁴	2	18.18
10 ⁴ -10 ⁶	4	36.36
10 ⁶ -10 ⁸	3	27.28
Total	11	100.00

Table (6): Statistical Analytical Results of Microbiological Examination of examined market yoghurt Samples based on their Coliform MPN count/gm.

Type of samples	No. of samples	+ve Samples	Min	Max	Mean	S.E.M ±
Market plain yoghurt	6	4	10x10 ³	20x10 ⁷	6.72x10 ⁶	6.66x10 ⁶
Market yoghurt fortified with Guava	6	3	94x10 ²	10x10 ⁹	7.31x10 ⁸	4.55x10 ⁸
Market yoghurt fortified with Mango	6	5	8.0x10 ²	3.3x10 ⁸	2.44x10 ⁷	1.20x10 ⁷
Market yoghurt fortified with Strawberry	6	4	3.3x10 ²	1.4x10 ⁹	8.85x10 ⁷	5.17x10 ⁷

Table (7): Frequency distribution of examined Market plain yoghurt samples based on their coliform count

Intervals	No of positive samples	%
10 ² -10 ⁴	3	15.78
10 ⁴ -10 ⁶	5	26.32
10 ⁶ -10 ⁸	5	26.32
10 ⁸ -10 ¹⁰	3	15.78
Total	16	100

Table (8): Statistical Analytical Results of Microbiological Examination of examined juice Samples based on their Coliform MPN count/gm.

Type of samples	No. of samples	+ve Samples	Min	Max	Mean	S.E.M ±
Guava Juice	6	4	10	30x10 ³	57.97x10 ²	19.38x10 ²
Mango Juice	6	3	10	90x10 ³	84.84x10 ²	42.64x10 ²
Strawberry Juice	6	6	20	15x10 ³	19.11x10 ²	6.19x10 ²

Table (9): Frequency distribution of examined Juice samples based on their coliform count/gm.

Intervals	No. of positive samples	%
10-10 ²	5	38.46
10 ² -10 ³	4	30.77
10 ³ -10 ⁴	4	30.77
Total	13	100.00

Table (10): Incidence of Mold & yeast in examined lab made yoghurt samples

Type of samples	No of Samples	Positive samples	%
Lab made plain yoghurt	6	2	33.33
Lab made yoghurt fortified with Guava	6	2	33.33
Lab made yoghurt fortified with Mango	6	3	50.00
Lab made yoghurt fortified with Strawberry	6	1	16.66

Table (11): Statistical Analytical Results of Microbiological Examination of examined lab made yoghurt Samples based on their Mold & yeast count/gm.

Type of samples	No of Samples	+ve Samples	Min	Max	Mean	S.E.M ±
Lab made plain yoghurt	6	2	100	10x10 ³	10x10 ²	5x10 ²
Lab made yoghurt fortified with Guava	6	2	100	75x10 ³	8.25x10 ³	2.69x10 ³
Lab made yoghurt fortified with Mango	6	3	50	176x10 ²	10x10 ²	6.34x10 ²
Lab made yoghurt fortified with Strawberry	6	1	100	6x10 ⁵	21.89x10 ³	12.84x10 ³

Table (12): Frequency distribution of examined Lab made yoghurt samples based on their Mold & yeast count/gm.

Intervals	No of positive samples	%
10-10 ²	1	12.50
10 ² -10 ³	4	50.00
10 ³ -10 ⁴	3	37.50
Total	8	100.00

Table (13): Incidence of Mold & yeast in examined market yoghurt samples

Type of samples	No of Samples	Positive Samples	%
Market plain yoghurt	6	3	50.00
Market fortified yoghurt by Guava	6	3	50.00
Market fortified yoghurt by Mango	6	2	33.33
Market fortified yoghurt by Strawberry	6	2	33.33

Table (14): Statistical Analytical Results of Microbiological Examination of examined market yoghurt Samples based on their Mold & yeast/count/gm.

Type of samples	No. of samples	+ve Samples	Min	Max	Mean	S.E.M ±
Market plain yoghurt	6	3	10x10 ³	20x10 ⁷	.72x10 ⁶	6.66x10 ⁶
Market yoghurt fortified with Guava	6	3	11x10 ²	74x10 ⁹	92.35x10 ⁴	21.16x10 ⁴
Market yoghurt fortified with Mango	6	2	77 x10 ⁴	27x10 ⁸	13.66x10 ⁶	57.27x10 ⁶
Market yoghurt fortified with Strawberry	6	2	83x10 ⁴	79x10 ⁷	21.43x10 ⁷	10.37x10 ⁷

Table (15): Frequency distribution of examined Market plain Yoghurt samples based on their Mold & yeast count/gm.

Intervals	No of positive samples	%
10^2-10^4	1	10.00
10^4-10^6	3	30.00
10^6-10^8	4	40.00
10^8-10^{10}	2	20.00
Total	10	100

DISCUSSION

Data represented in Tables (1&2) reported that, coliforms were detected in 33.33%, 66.76%, 50.00% and 33.33% of examined Lab made plain yoghurt and Lab made fortified yoghurt (Guava, Mango and Strawberry juice) respectively. On the other hand coliforms were present in market plain yoghurt in 66.66.00% of the examined samples, while in market fortified yoghurt with Guava, Mango and Strawberry the incidence of coliforms was 50.00%, 83.33 % and 66.66% respectively.

The prevalence of Coliforms was illustrated in Table (3) from which it was clear that Coliforms was present in 66.76 %, 50.00% and 33.33% of examined Guava, Mango and Strawberry juice samples respectively.

Nearly similar findings were reported by Saudi(1980); Abeer(1997) and Hanaa(1999). Lower findings were recorded by Lopez *et al.*(1993); Shahid *et al.*(2002); Zakai & Erdogan (2003) and Riadh Al Tahiri (2005), while high counts were reported by Hafez (1984); Ayoup(1986) and Aboubaker (2004).

It is clear from the obtained results that all the examined yoghurt samples were positive to coliforms and not agree with the Egyptian Standard Specification (2005) which Recommended that coliforms count should be less than 10 cells /gm in the product.

Coliforms are Gram negative spores forming rods, facultative anaerobes resistant to many surface agents, and ferment lactose to produce lactic acid and gas within 48 hours at 32 or 35co. Coliforms are represented by four genera of the family Enterobacteriaceae: Citrobacter, Enterobacter, Escherichia and Klebsiella. Coliforms are able to grow in pH range from 4.4 to 9 (few of them are resistant to acid food).

Lucea (1995) mentioned that coliforms will be unable to survive at low pH in yoghurt and this inhibition is reinforced by the production of antibiotic substances which is produced by the yoghurt starter.

High coliforms count in dairy products render the product of inferior quality and cause economic losses (ICMSF,1980).Coliform tests for dairy products are not intended only to indicate fecal contamination but do reflect over all dairy farms and plant sanitation (Reinbold,1983).

Coliforms are proven to use as safety indicator so used as a component of safety programs such as HACCP system. The presence of coliform in food especially heat-processed foods is probably due to improper sanitation after heat treatment (Ray, 2004), contamination with fecal matter and their presence related to presence of enteric pathogen.

Results recorded in Table (4) showed that the minimum, the maximum and the mean of coliforms in lab made plain yoghurt were 10×10^3 , 9.8×10^6 and $6.35 \times 10^5 \pm 2.95 \times 10^5$ /gm. While the mean value of coliforms in fortified yoghurt with Guava, Mango and Strawberry juice were $6.35 \times 10^4 \pm 2.78 \times 10^4$; $2.71 \times 10^6 \pm 1.32 \times 10^6$ and $13.06 \times 10^5 \pm 6.90 \times 10^5$ /gm in examined yoghurt samples respectively. The high frequency (36.36%) lied within the range 10⁴-10⁶ (Table 5).

Inspection of Table (6) showed that the minimum coliforms in market plain yoghurt respectively was 10×10^3 , the maximum was 20×10^7 and the mean was $6.72 \times 10^6 \pm 6.66 \times 10^6$ /gm.

As regarded here in this study and recorded in Table (6), it is clear that the minimum coliform content in fortified market yoghurt with guava, mango and Strawberry were 94×10^2 , 8.0×10^2 and 3.3×10^2 ; while the maximum were 10×10^9 , 3.3×10^9 and 1.4×10^9 respectively with a mean average of $7.31 \times 10^8 \pm 4.55 \times 10^8$, $2.44 \times 10^7 \pm 1.20 \times 10^7$ and $8.85 \times 10^7 \pm 5.17 \times 10^7$ /gm respectively.

The findings in Table (7) display the frequency distribution of coliform count and show that the highest frequency distribution of coliform count per gm of market plain yoghurt (26.23%) lies within the range (10⁴-10⁶) and (10⁶-10⁸).

Figures tabulated in Table (8) show that the minimum and maximum coliform counts / gm. of examined guava, mango and strawberry juice samples were 10, 30×10^3 ; $10,90 \times 10^3$ and $20,15 \times 10^3$, with a mean value of $57.97 \times 10^2 \pm 19.38 \times 10^2$, $84.84 \times 10^2 \pm 42.64 \times 10^2$ and $19.11 \times 10^2 \pm 6.19 \times 10^2$ in examined juice samples respectively. The high frequency distribution of coliform (38.46%) lies within the range 10¹-10² (Table 9).

Inspection of Table (10) revealed that 2 (33.33%); 2 (33.33%); 3 (50.00) and 1 (16.66%) of lab plain and fortified (Guava, mango and strawberry) yoghurt examined samples contained molds and yeast respectively.

It is clear from the results given in Tables (11) and that the minimum, maximum and mean mold & yeast count / gm of lab made and fortified (guava, mango and strawberry) yoghurt samples were (100,100,50 and 100); (10×10^3 , 75×10^3 , 176×10^2 and 6×10^5); ($10 \times 10^2 \pm 5 \times 10^2$, $8.25 \times 10^3 \pm 2.69 \times 10^3$, $10 \times 10^2 \pm 6.34 \times 10^2$ and $21.89 \times 10^3 \pm 12.84 \times 10^3$) respectively.

The highest frequency (50.00 %) lies within the range 10²-10³ (Table 12). Table (13) show that 3 (50.00%); 3 (50.00%); 2 (33.33) and 2 (33.33%) of market plain and fortified (Guava, mango and strawberry) yoghurt examined samples contained molds and yeast respectively.

It is clear from the data obtained in Tables (14) that the minimum, maximum and mean mold & yeast count / gm of market plain and fortified (guava, mango and strawberry) yoghurt samples were (10¹, 11¹, 77¹ x10⁴ and 83¹ x10⁴); (20×10^7 , 74×10^5 , 27×10^8 and 79×10^7); ($6.72 \times 10^6 \pm 6.66 \times 10^6$, $92.35 \times 10^4 \pm 21.16 \times 10^4$, $13.66 \times 10^6 \pm 57.27 \times 10^6$ and $21.43 \times 10^7 \pm 10.37 \times 10^7$) respectively. The majority of the samples (40%) were lies within the range 10⁶-10⁸ (Table 15).

Nearly similar findings were reported by Rodriguez- Ferri *et al.* (1978); Mansour (1986); Abeer (1997) and Hanaa (1999), while higher findings were reported by Uden and Sousa (1957) lower values were recorded by Aarnott *et al.* (1974); L alas (1985) and Lopez, *et al.* (1993), all contaminated samples were out of the Egyptian standard specifications (2005) such stated that the fungal count must be ≤ 10 cell/g. and with permissible limit of mycotoxins.

The high contamination level with yeasts and moulds in the samples of balady yoghurt indicates neglected hygienic measures during production, handling and distribution of such product. Abou Donia (1980) attributed the contamination of yoghurt with yeast & mould in Egypt to post production contamination. Con *et al.* (1996) mentioned that the high contamination level within yoghurt examined samples was due to contamination from air day old culture used for yoghurt manufacture and agreed with Yaygm and Kilic (1980) who showed that yoghurt made from pure culture has no growth of yeast and mould up to 4 days the storage.

The main microbiological problems associated with yoghurt, juice, blends of yoghurt juice drinks, is the spoilage caused by yeast and mould (Garbutt *et al.* 1997) yeast are very common in yoghurt, juice, blends of yoghurt juice drinks, while the mould are less problem than yeast (Robinson, 1990 and Pitt & Hcking, 1997) Alekiewa and Mirkov (1979) found that 3.5% of the yoghurt lots presented for sale on markets contained yeast, while one lot had mould. Li and Li (1998) recorded that 56.67% of examined yoghurt samples were contaminated with yeast but of 67.33% of the examined samples which were contained with yeast and mould.

Yeast contaminated the yoghurt and their products causing economic losses undesirable changes such as frothy consistency and yeasty flavour. Moreover, some species of yeasts constitute a public hazard such as gastrointestinal disturbance, endocarditis, and occasionally fatal systemic diseases (Marth *et al.* 1972 and Jaquet & Teherani, 1976).

Although the presence of mould in yoghurt constitute a serious economic losses because it associated with a visible spoilage, off flavor, discoloration and rejection of the product but also isolation of some species have raised the possibility that contaminated yoghurt could be source of mycotoxins which were implicated in outbreaks of human food poisoning and many several diseases such leukemia, cancer and kidney toxicity (Bullerman, 1981, and Robinson, 1990).

CONCLUSION

From our survey in the present study, allow concluding that there are 4 groups of samples of Yoghurt (lab made yoghurt (Balady) market), Yoghurt fortified with juice, Market yoghurt fortified by juice and juice (Guava, Mango, Strawberry). Especially yoghurt (Lab made & Market) are of inferior quality and subjected to many risks of contamination due to neglected hygienic and sanitary measures adopted during production, handling, transportation and marketing, especially lab made yoghurt which depend upon traditional system. Such system could pose favorable environment for bacterial

contamination and multiplication. The unclean hands of worker, poor quality of milk used, unhygienic conditions of manufacturing unit, inferior quality of materials used and water supplied for washing utensils, could be the source of accelerating the bacterial contamination and the post- manufacturing contamination of these products. Lack of proper cooling storage with ambient summer temperature of Egypt is also factors magnitude of the problem of bacterial contamination.

The objectionable contamination of the examined samples with different types of coliforms and enterococci, as well as other contaminants is never desirable and may be responsible for loss quality and spoilage of the products that render them at times inedible. Moreover, the presence of pathogenic organisms in these products reveals that commercial's yoghurts may at times constitute a public health hazard. From previous results indicate that all groups of samples E.coli is negative; that in plain yoghurt (balady & market) coliform count at zero time is (2.95x10⁵), (6.66x10⁶) and after 2 weeks for storage at 4⁰C ; At yeast & Mould is(5x10²) ,(6.66x10⁶). In yoghurt (Lab made & Market) fortified with juice (Guava, Mango, Strawberry) coliform count at zero time is (2.78x10⁴, , 1.32x10⁶, 6.90x10⁵), (4.55x10⁸ , 1.20x10⁷ , 5.17x10⁷) and after 2 weeks for storage at 40C ; and at yeast & Mould are(2.69x10³ , 6.34x10² , 12.84x10³) , (21.16x10⁴, 57.00x10⁶, 9.35x10⁷) .

So from last results the best samples which growing of samples decreasing or prevented in (Lab made plain yoghurt, Market fortified yoghurt by Guava, Strawberry Juice).

Therefore, to safe-guard consumers from being infected and after storage at 4⁰C for two weeks for yoghurt and 6months for juice, to save a lot of the products from being spoiled on the market, more elaborative measures from the point of production of yoghurt, juice, and blends of yoghurt juice drinks to the point of consumption and at all intermediary levels are required:

The dairy processing plant from which the yoghurt, juice, and blends of yoghurt juice drinks originate, is approved, licensed and monitored by the competent state Regulatory Agency and is producing yoghurt, juice, and blends of yoghurt juice drinks according to the Federal laws and regulations. Good quality, safe yoghurt, juice, and blends of yoghurt juice drinks under strict hygienic conditions. Licenses should be given to establishments after all equipment, facilities and hygienic conditions are fulfilled. Implantation of HACCP plan built upon a solid foundation of prerequisites programs is required for each product for the safety production of such products.

In conclusion, it seems necessary that concerned authorities should impose regulations and bacteriological standards for yoghurt, juice, and blends of yoghurt juice drinks, taking active part in the control of yoghurt, juice, and blends of yoghurt juice drinks production and handling as well as improving the quality of produced yoghurt, juice, and blends of yoghurt juice drinks.

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تقييم الجودة الميكروبية في الحليب والزبادى المخلوطين ببعض الفواكه وعصائرها
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***** قسم الصناعات الغذائية - المركز القومى للبحوث - مصر**

هذه الدراسة توضح أن اللبن الرائب العادى يصنع بحجم انتاج المعمل من ألبان الأبقار المؤخوذة من بعض المزارع المعتمدة من القاهرة /مصر . اللبن الرائب ممزوج مع عصائر طازجة وهى (عصير الجوافة- عصير المانجو- عصير الفراولة) وأيضا يتم دراسة اللبن الرائب تجاريا ممزوج مع عصائر طازجة . نوعية المواد الكيميائية والجرثومية للبن الرائب ، اللبن الرائب الممزوج بالعصير ، العصير الطازج حققت خلال فترة التخزين داخل المبردة عند درجة حرارة 4 درجة مئوية لمدة أسبوعين اللبن الرائب وستة أشهر للعصير . التحليل الجرثومى يوضح تأثير نمو الخميرة والعفن، وكوليفورم قبل وبعد التخزين . ونتيجة لهذه الدراسة توضح أن العصير الطازج له تأثير كبير على قبول اللبن الرائب قبل وبعد التخزين .

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