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# FORTIFICATION OF PROBIOTIC STIRRED YOGHURT BY ADDITION OF APPLE AND MANGO PULPS

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**ABSTRACT:** Apple (*Malus domestica*) and mango (*Mangifera indica* L.) pulps were used as a prebiotic in the fortification of flavoured probiotic stirred yoghurt. Flavoured probiotic stirred yoghurt was made from cow's milk supplemented with apple and mango pulps at levels of 5.0, 10.0 and 15%. Resultant stirred yoghurt of all treatments was stored at 5<sup>°</sup>C for 15 days to study the physico-chemical, microbiological, organoleptic and colour properties (L<sup>\*</sup>, a<sup>\*</sup>, b<sup>\*</sup>) of yoghurt samples during storage period. Addition of apple and mango pulps to cow's milk increased acidity, total solids, but decreased total protein, total fat content and pH values of resultant flavoured probiotic stirred yoghurt. Also apple and mango pulps addition improved starter activity, increased total bacterial, bifidobacterial and lactobacilli bacterial counts of resultant flavoured probiotic stirred yoghurt. Colour characteristics (L<sup>\*</sup>, a<sup>\*</sup>, b<sup>\*</sup>) are varied according to the kinds of fruit pulp used in yoghurt, which caused changes in colour of the yoghurt samples. These observations were associated with the level of mango and apple pulp addition. As a result of the organoleptic evaluations, 10% apple pulp yoghurt sample had the highest overall acceptability. Yoghurt with 10% fruit pulp, especially apple pulp could be recommended for large scale production of fruit probiotic yoghurt.

Key words: Apple and mango pulps, fortification, probiotic, stirred yoghurt, prebiotic, cow's milk.

## **INTRODUCTION**

According to the Code of Federal Regulations of the United States Food and Drug Administration (FDA, 2013), yoghurt can be defined as a food produced by culturing one or more of the optional dairy ingredients namely, cream, milk, partially skimmed milk, and skim milk, used alone or in combination with a characteristic bacterial culture that contains lactic acid producing bacteria, *Lactobacillus bulgaricus* and *Streptococcus thermophilus*.

On the other hand, the bio- or probiotic yoghurt refers to those made using cultures of beneficial microorganisms that claim to have numerous health benefits once ingested, typically the probiotic strains of bifidobacteria and *L. acidophilus*. These probiotic strains are said to claim more specific health benefits and represent the types of friendly microflora present in the gut (**Dowden, 2013**).

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In recent years, many consumers have developed an interest in learning more about nutrition and food. Health, convenience, and private labels have become key drivers of the world's fastest-growing food and beverage categories. Consumers want food that is inherently healthy, yet easy to prepare and consume, especially with women and men working an average of 7 and 8 hr per day, respectively (**Sloan, 2005**).

Functional foods can come in many forms, some of which are conventional foods, such as yoghurt, with bioactive components inherent to the food. Others may be foods that are specifically enhanced to reduce the risk of disease, such as yoghurt beverages fortified with probiotics/prebiotics to maintain the health of the colon. More research is needed to identify the role of bioactive components such as prebiotics to determine the optimal levels for the population subgroups and the best vehicle foods to be enhanced to target specific population subgroups (IFT, 2006). According to Codex Alimentarius Standard (2003), to provide health benefits, the concentration of probiotic bacteria should be not less than  $10^6$  CFU/g of the product.

The World Health Organization (WHO, 2005) recommends consumption of fruits and vegetables as part of a healthy diet with daily recommendation of 5 servings or at least 400 g per day. Fruits and vegetables are good sources of vitamins, minerals, antioxidants and fibres. So, certain fruits can be used in yoghurt production for improving their nutritional values and sensory properties "*e.g.* strawberry, apple, cornelian, rosehip, morello cherry, grape, date and other fruit homogenates" (Erdogan and Zekai, 2003).

Addition of different fruits in yoghurt manufacture has been attempted increasingly. Use of fruit in yoghurt makes it more delicious. This product contains both the refreshing flavour of fruit and beneficial effect of yoghurt. Fruit yoghurt has more taste and pleasing flavour (**Mahmood** *et al.*, 2008).

Introduction of various fruit-flavoured yoghurts has significantly contributed to the consumption of yoghurt from all ages. Fruits may be added to yoghurt formulae as single or blends in the form of refrigerated, frozen, canned fruit, juice or syrup. Most common fruits used in yoghurt formula are peach, cherry, orange, lemons, purple plum, boysen berry, spiced apple, apricot, pineapple, strawberry, raspberry and blueberry (Chandan and Shahani, 1993).

Fruit mixes improve the nutritional value and the taste of yoghurt, and fruit enhancement plays a considerable role in yoghurt consumption and sales. Varieties of fruit yoghurts have been formulated as probiotic fruit yoghurts, and the survival of probiotic bacteria in yoghurts has been investigated during cold storage (Kailasapathy *et al.*, 2008).

Results of recent studies indicated that fruit addition levels plays a significant role in acceptability of fruit added yoghurt. A study of the higher level of fruit addition into yoghurt would be increase sensory quality of the fruit yoghurts (Tarakçı and Küçüköner, 2003a).

Thus, the objective of this study was to produce probiotic stirred yoghurt enriched with different levels of apple and mango pulps. Effect of these additives on the physicochemical, microbial, sensory qualities and overall acceptability of the product was also a goal of the study.

## **MATERIALS AND METHODS**

## **Materials**

## Milk

Fresh cow's milk was obtained from Faculty of Veterinary Medicine farm, Zagazig University, Egypt.

#### Skim milk powder

Skim milk powder was purchased from private dairy plant in Zagazig, Egypt.

### Table sugar

Commercial grade clean, white crystalline cane-sugar was obtained from local market.

## **Yoghurt Cultures**

ABT-5 culture (Lactobacillus acidophilus; Bifidobacterium bifidium and Streptococcus thermophilus) and ABT-3 culture (Lactobacillus acidophilus LA-5, Bifidobacterium animalis subsp. lactis BB-12, and Streptococcus salivarius subsp. thermophilus) were obtained from Chr. Hansen Holding A/S, Boege Alle 10-12, 2970 Hoersholm, Danmark.

Streptococcus thermophillus (EMCC1044 = DSM20479) and Lactobacillus delbrueckii ssp. bulgaricus (EMCC1102 = DSM20080) were used as a yoghurt starter. These strains were obtained from Microbiological Resources Center (Cairo MIRCEN), Egyptian Microbial culture collection (EMCC), Faculty of Agriculture, Ain Shams University, Egypt.

Streptococcus thermophilus was revived by a series of two inoculations into 10 ml of  $M_{17}$  broth and incubated aerobically at  $37^{\circ}$ C for 48 hr., and *L. delbrueckii* ssp. *bulgaricus* was inoculated into MRS broth and incubated at

42°C for 48 hr., anaerobic chamber (**Dave and Shah**, 1996).

The stock cultures were maintained at -80°C in 12% (*W/W*) reconstituted skim milk (RSM) and 40% (*V/V*) steriled glycerol. The microorganisms were activated by growing in 10% (*W/W*) sterile RSM for 18 hr., consequently 3 times prior to yoghurt manufacture.

#### **Preparation of fruit pulp**

Fresh ripe fruits (Golden Delicious apple and Keitt mango) were gentility washed under tap water, peeled with the help of a knife aseptically and the seeds were removed manually then, the cut pieces were quickly mashed using an electric mixer (Braun, Germany) and finally converted into homogenous apple and mango pulp and sweetened with cane sugar at level of 6%. Homogenized fruit pulps were then blanched at  $80 \pm 1^{\circ}$ C for 5 minutes. After cooling, pulps were kept at refrigeration temperature (4°C) in sterilized PET (Polyethylene terephthalate) bottle until the preparation of yoghurt.

## Preparation of flavoured probiotic stirred yoghurt

Fresh whole cow's milk, was fortified with 2% skim milk powder to increase its solids. Milk was heated at 90°C for 15 mint and then rapidly cooled to 40°C. Milk was divided into equal 7 portions. Milk of all treatments was then inoculated with 2% of voghurt culture and 2% mixed probiotic culture containing 1:1 ratio of ABT-5 and ABT-3 cultures. Then incubated at 40°C until complete coagulation (about 4 hours) and cooled overnight. The first portion was stirred without additives as a control. The calculated amount of pasteurized apple and mango pulps (5.0, 10.0 and 15%) were stirred with the other 6 portions, respectively to obtain stirred flavoured yoghurt. The mixture was mixed with electric stirrer to obtain stirred yoghurt. The resultant product of all treatments was packed into sterilized plastic cups (100 ml) and stored at 5°C for 15 days. Samples for chemical, microbiological analyses and sensory evaluation were taken after 0, 5, 10 and 15 days. The different treatments could be summarized as follows:

C: Bio stirred yoghurt (control)

A<sub>1</sub>: Bio stirred yoghurt + 6 percent sugar +5 percent apple pulp

A<sub>2</sub>: Bio stirred yoghurt + 6 percent sugar +10 percent apple pulp

A<sub>3</sub>: Bio stirred yoghurt + 6 percent sugar +15 percent apple pulp

**M<sub>1</sub>:** Bio stirred yoghurt + 6 percent sugar +5 percent mango pulp

**M<sub>2</sub>:** Bio stirred yoghurt + 6 percent sugar +10 percent mango pulp

**M<sub>3</sub>:** Bio stirred yoghurt + 6 percent sugar +15 percent mango pulp

#### **Methods of Analyses**

Resultant stirred yoghurt from all treatments was chemically analyzed for total solids and fat according to **AOAC** (2010). Total protein per cent was determined by semi-micro Kjeldahl method according to **AOAC** (2010).

#### **Rheological Measurements**

#### Syneresis

The released whey from yoghurt samples was measured according to the method of **Aryana (2003)**. The quantity of whey collected from every sample in a graduated cylinder after 2 hr., of drainage at 20 °C was used as index of syneresis.

#### Viscosity

Viscosity of stirred yoghurt samples was determined by the method of **Aryana (2003)** using Rotational Viscometer Type Lab. Line Model 5437. Results were expressed as CPS.

#### **Colour analyses**

A Hunter colorimeter (Hunter Laboratories, Reston, VA, USA) was used to determine of Hunter  $L^*$  (black to white),  $a^*$  (green to red) and  $b^*$  (blue to yellow) color parameters of yoghurt samples.

#### **Microbiological Analyses**

#### **Enumeration of bifidobacteria**

The MRS-NNLP (Nalidixic (0.030g), neomycin sulfate (0.200g), lithium chloride (0.600 g) and paromomycin sulfate (0.250 g) medium (Oxoid, 2006) was prepared according to the method described by Laroia and Martin (1991). The basal medium was MRS agar. Filtersterilized NNLP was added to the autoclaved MRS base just before pouring. Filter-sterilized L-cysteine-HCl (0.05% final concentration) was also added at the same time to lower the oxidation reduction potential of the medium and to enhance the growth of anaerobic bifidobacteria.

#### Enumeration of lactobacilli strain

MRS agar (Oxoid, 2006) with pH 6.2±0.1 was used for enumeration of Lactobacillus delbrueckisub ssp bulgaricus according to Dave and Shah (1996). The plates were incubated at 42°C for 72 hr. Anaerobic culture jars (2.5 L) employed generate anaerobic were to conditions, atmospheric oxygen being absorbed by means of (AnaeroGen AN 25 sachets Oxoid). The counts were expressed as cfu/ml. The lactobacilli identified on the basis of colonial which confirmed by microscopic type examination. Lactobacillus was Gram positive rods with rounded ends.

#### Enumeration of coliform bacteria

Total coliform count was estimated by plating suitable dilution on Violet Red Bile Agar medium (VRBA) (**Oxoid, 2006**) as described by American Public Health Association **APHA** (2004). The plates were incubated at  $35^{\circ}$ C for 24 hr., and the small non-mucous red colonies were counted.

#### Enumeration of moulds and yeasts

Moulds and yeasts were enumerated on acidified potato dextrose agar medium (Difco, 2003). Plates were incubated at  $25^{\circ}$ C for 4-5 days.

## **Sensory evaluation**

The sensory evaluation for the flavoured probiotic stirred yoghurt samples was done by trained panelists from the staff members of Food Science Department, Faculty of Agriculture, Zagazig University according to the method as reported by **Hamdy** *et al.* (1972). The score was rated on hedonic scale of 10 for appearance/ colour, 45 for taste/flavour, 35 for body/texture and 10 for acidity.

## **RESULTS AND DISCUSSION**

## **Physicochemical Properties of Fruit Pulps**

Physicochemical properties of apple and mango pulps used for flavoured probiotic stirred yoghurt making are shown in Table 1. Results showed that, apple pulp had higher moisture, carbohydrates, fat and total soluble solids compared with mango pulp and lower acidity, protein, fibres, ash and total solids. These values are consistent with values reported by **Campeanu (2009) and Appiah** *et al.* (2011).

#### Acidity and pH

Mean of titratable acidity and pH of control and flavoured probiotic stirred yoghurts are presented in Table 2. The variation in pH and acidity of yoghurts were differed due to the effects of different fruit pulps. Addition of fruit pulp decreased the pH in yoghurt. Whereas, titratable acidity increased with increasing fruit pulp percentage. The increase in acidity may be due to that apple and mango pulps contain higher acidity than milk. This increase in flavoured voghurt acidity was associated with a decrease in pH values of resultant product. Moreover, acidity contents of yoghurt of all treatments slightly increased during storage period and this was associated with a gradual decrease in pH values. This might due to the growth and activity of microorganisms. The general trend of these results is in agreement with those reported by Roy et al. (2015). The result of this study indicates that, 15% mango flavoured yoghurt had the highest titratable acidity, but it had the lowest pH (4.71).

# Chemical Analysis of Flavoured Probiotic Stirred Yoghurt

The results of chemical analyses of yoghurt samples are presented in Table 3. Control sample had the highest moisture content (89.63%) while the flavoured yoghurt sample containing 15% apple pulp had the lowest moisture content (82.22%). This showed that moisture levels decreased with increase in the concentration of the apple and mango pulps added. With increasing of apple and mango pulps levels to cow's milk, total solids (TS) content was raised. This was associated with a

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Type of pulp	Moisture	Total solids	TSS	Protein	Fat	Fibres	Carbohydrates	Ash	Acidity
Apple pulp	84.80	15.20	13.10	0.20	0.80	0.30	11.9	0.29	0.28
Mango pulp	83.10	16.90	12.60	0.70	0.53	1.02	9.73	0.43	0.47

 Table 1. Physicochemical properties of apple and mango pulps (%)

Table 2. Acidity and pH of flavoured probiotic stirred yoghurt during storage period at 5°C for 15 days

Yoghurt	Titrat	able acidity	(as lactic ad	cid %)	рН								
sample -			e period ay)		Storage period (day)								
	0	5	10	15	0	5	10	15					
С	0.88	0.94	1.01	1.06	4.76	4.63	4.56	4.51					
$A_1$	0.90	0.96	1.03	1.08	4.75	4.62	4.45	4.40					
$A_2$	0.92	0.96	1.02	1.08	4.75	4.65	4.43	4.39					
A <sub>3</sub>	0.92	0.98	1.04	1.09	4.74	4.63	4.41	4.35					
$M_1$	0.94	1.00	1.06	1.11	4.73	4.63	4.51	4.47					
M <sub>2</sub>	0.96	1.02	1.09	1.10	4.72	4.59	4.42	4.37					
M <sub>3</sub>	0.98	1.04	1.10	1.15	4.71	4.61	4.49	4.45					

C: Bio stirred yoghurt (control)

 $A_1$ : Bio stirred yoghurt + 6 per cent sugar +5 per cent apple pulp

A<sub>2</sub>: Bio stirred yoghurt + 6 per cent sugar +10 per cent apple pulp

 $A_3$ : Bio stirred yoghurt + 6 per cent sugar +15 per cent apple pulp

 $M_1$ : Bio stirred yoghurt + 6 per cent sugar +5 percent mango pulp

**M**<sub>2</sub>: Bio stirred yoghurt + 6 per cent sugar +10 per cent mango pulp

 $M_3$ : Bio stirred yoghurt + 6 per cent sugar +15 per cent mango pulp

Table 3. Chemical analysis of flavoured probiotic stirred yoghurt during storage period at 5°C for 15 days

Yoghurt		Chemical properties in percentage														
sample		Moi	sture		<b>Total solids</b>					al fat		Total protein				
	5	0	e perio ay)	d	Storage period (day)				Storage period (day)				Storage period (day)			
	0	5	10	15	0	5	10	15	0	5	10	15	0	5	10	15
С	89.63	89.57	89.43	89.34	10.37	10.43	10.57	10.66	4.20	4.40	4.60	4.70	4.80	4.96	5.18	5.35
$\mathbf{A}_{1}$	86.73	86.29	86.59	86.52	13.27	13.71	13.41	13.48	4.10	4.30	4.40	4.60	3.95	4.25	4.41	4.54
$\mathbf{A}_{2}$	82.26	82.11	82.03	81.84	17.74	17.89	17.97	18.16	4.00	4.20	4.30	4.50	3.82	4.10	4.25	4.37
$A_3$	82.22	81.81	81.63	81.31	17.78	18.19	18.37	18.69	3.90	4.10	4.20	4.40	3.49	3.60	3.75	3.78
$\mathbf{M}_{1}$	86.19	85.81	85.60	85.51	13.81	14.19	14.40	14.49	3.80	3.90	4.00	4.20	4.77	5.04	5.18	5.24
$M_2$	84.62	84.23	84.00	83.70	15.38	15.77	16.00	16.30	3.70	3.80	4.00	4.10	4.48	4.63	4.84	4.92
M <sub>3</sub>	84.34	84.18	83.90	83.55	15.66	15.82	16.10	16.45	3.60	3.80	3.90	4.00	4.33	4.58	4.70	4.75

For Note see: Table 2.

slight decrease in fat and protein contents. The increase in total solids could be due to the highest total solids of apple and mango pulps. The results for total solids content were similar to those obtained by Fadela et al. (2009). The fat content was decreased from 4.20 (C) to 3.60  $(M_3)$ . The decrease in fat content could be due to apple and mango pulp contains less fat than milk. The results for fat content were similar to those obtained by Hossain et al. (2012a). The results of this study indicate that, the maximum and minimum protein content was found in control and yoghurt containing 15% apple pulp, respectively. The decrease in protein content may be due to addition of varied proportion of apple and mango pulps in yoghurt drink and as the protein content of the apple and mango pulp is less than milk. The results for protein content were similar to those obtained by Osundahunsi et al. (2007) and Hossain et al. (2012b).

## Rheological Properties of Flavoured Probiotic Stirred Yoghurt

Syneresis is the losing out of liquid from yoghurt. Syneresis is one of the key quality parameters for yoghurt. Higher level of syneresis shows that yoghurt is of low quality. The syneresis of yoghurts were affected by both fruit concentration and storage time and the changes are shown in Table 4. Syneresis values of different types of voghurt varied from 19.00 to 32.50 per cent. These results are matched with Tarakçı and Küçüköner (2003a). The addition of fruit pulp caused a decrease of syneresis in all samples of yoghurts. It could be related to the capacity to absorb water by solid and fibres that present in fruit which leads to a decrease of syneresis (Mahmood et al., 2008). The viscosity of yoghurt samples decreased slowly at the 10<sup>th</sup> day and continued to decrease rapidly at the 15<sup>th</sup> day of storage as showen in Table 4. Flavoured stirred yoghurt with 15% mango pulp has the highest viscosity (488.00 CP). In general, a substantial reduction in viscosity occurred during the storage of yoghurt samples. The viscosity of yoghurt increases with added apple and mango pulp. The viscosity generally varies depending on the amount of fruit in yoghurt (Akin and Konar, 1999). These results are matched with Arslan and Ozel (2012).

#### **Colour Properties**

Comparing the colorimetric parameters of flavoured probiotic stirred yoghurt is shown in Table 5. Apple and mango pulps added to the yoghurt samples had a strong effect on the colour properties. Colour measurement showed the slight difference of colour between different kinds of yoghurt samples. With the addition of apple and mango pulps, L<sup>\*</sup> values of yoghurt samples were decreased while a<sup>\*</sup> and b<sup>\*</sup> values were increased. In the case of all yoghurt samples, the L<sup>\*</sup> and b<sup>\*</sup> values were observed to be very high throughout the storage period indicating that the product retained the appealing whiteness while a<sup>\*</sup> value decreased during the storage period in yoghurt samples as a result of the decline in yoghurt syneresis because the whey released by the gel contains riboflavin, which has a very important green component. In several studies, depending on the colours of the fruit and other additives added in yoghurt, the colour properties  $(L^*, a^*, b^*)$  of voghurt samples changed. These results in general are similar to those obtained by Bartoo and Badrie (2005).

# Total bacterial count, bifidobacterial and lactobacilli bacterial count

Results in Table 6 present the total bacterial, bifidobacterial and lactobacilli bacterial counts of control and fruit flavoured yoghurt samples. Results showed that, total bacterial, lactobacilli bacterial, and bifidobacterial counts were affected by the type of fruit and their percentages in yoghurt. Total bacterial count was increased with increased of fruit pulp percentage in fruit flavoured yoghurt samples, which supported bacterial growth. Similar were reported by Tarakçı results and Küçüköner (2003b) who stated that, total bacterial count in control sample was lower than yoghurt samples containing fruit juice. The results of this study indicates that, among fruit flavoured yoghurt samples, the microbial quality of yoghurt containing 5% mango was least for total bacterial count (84 cfu  $\times$  10<sup>6</sup>/g), bifidobacterial count (35 cfu  $\times$  10<sup>6</sup>/g) and lactobacilli bacterial count (60 cfu  $\times$  10<sup>6</sup>/g). The bifidobacterial count was decreased with increasing storage period (Table 6). This may be

Yoghurt		Viscosity (	Centipoise)		Whey syneresis (ml/100 g)								
sample			e period ay)		Storage period (day)								
	0	5	5 10		0	5	10	15					
С	295.00	330.00	319.00	279.00	29.10	31.00	31.50	32.50					
$\mathbf{A}_{1}$	323.00	372.00	355.00	320.00	28.20	30.70	31.00	31.50					
$\mathbf{A}_{2}$	331.00	373.00	349.00	325.00	26.50	29.00	29.50	30.00					
$A_3$	362.00	400.00	387.00	350.00	25.50	26.00	27.50	28.50					
$\mathbf{M}_{1}$	426.00	454.00	409.00	393.00	24.00	25.70	26.50	27.50					
$M_2$	450.00	521.00	443.00	421.00	21.00	22.50	23.00	24.50					
$M_3$	488.00	536.00	458.00	430.00	19.00	21.00	22.00	23.00					

Table 4. Rheological properties of flavoured probiotic stirred yoghurt during storage period at  $5^\circ C$  for 15 days`

For Note see: Table 2.

Table 5. Colour parameters (L<sup>\*</sup>, a<sup>\*</sup>, b<sup>\*</sup>) of flavoured probiotic stirred yoghurt during storage period at 5°C for 15 days

Yoghurt		Hunter values		Hunter values After 15 days of storage							
sample		Fresh samples									
•	L <sup>*</sup> value	a <sup>*</sup> value	b <sup>*</sup> value	L <sup>*</sup> value	a <sup>*</sup> value	b <sup>*</sup> value					
С	84.90	- 4.17	12.51	85.70	- 6.21	15.68					
$\mathbf{A}_{1}$	79.92	- 3.47	13.58	83.42	- 4.96	16.92					
$\mathbf{A}_{2}$	78.77	- 2.07	15.33	80.33	- 2.95	17.63					
$A_3$	77.36	- 1.79	16.14	79.94	- 2. 47	18.30					
$\mathbf{M}_{1}$	80.77	- 3.17	20.06	83.07	- 4.46	23.27					
$M_2$	80.17	- 2.82	22.19	82.97	- 4.31	25.01					
$M_3$	79.14	- 2.47	24.11	81.56	- 3.71	26.36					

 $\mathbf{L}^{*}$  (Lightness),  $\mathbf{a}^{*}$  (Redness/greenness) and  $\mathbf{b}^{*}$  (Yellowness/blueness). For Note see: Table 2.

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Yoghurt sample	To		terial co 10 <sup>6</sup> /g)	ount	Bi	fidobact (cfu×	terial co 10 <sup>6</sup> /g)	ount	Lactobacilli bacterial count (cfu×10 <sup>6</sup> /g) Storage period (day)					
			e perioc ay)	1			e perioc ay)	1						
	0	5	10	15	0	5	10	15	0	5	10	15		
С	78	96	107	121	24	19	15	11	57	50	44	37		
$A_1$	111	138	205	265	51	45	40	35	117	110	98	95		
$A_2$	118	142	225	289	59	52	46	38	128	121	115	108		
$A_3$	146	165	260	310	63	55	48	41	134	125	118	111		
$M_1$	84	118	132	146	35	29	26	19	60	53	47	41		
$M_2$	90	124	147	169	43	36	32	25	77	68	60	52		
$M_3$	98	118	189	212	49	41	36	31	90	78	67	57		

Table 6. Total bacterial, bifidobacterial and lactobacilli bacterial counts of flavoured probiotic stirred yoghurt during storage period at 5°C for 15 days

For Note see: Table 2

due to decreasing the pH values which observed in Table 2. The optimum pH for the growth of bifidobacteria is 6-7 and no growth at pH 4.5-5 or below (**Shah**, 2006).

### **Moulds, Yeasts and Coliforms Counts**

Results in Table 7 show the counts of yeasts, moulds and coliforms of flavoured yoghurt. Results indicated that flavoured yoghurt had higher yeasts count either fresh or after 15 days of storage. This could be due to the higher sugar content of fruit pulp which may enhance yeasts growth. Also it could be noticed that moulds and coliforms counts in fresh flavoured yoghurt samples were not detected and were some slight higher than control one after 15 days of storage in samples of all treatments. Meanwhile counts of yeasts, moulds and coliforms in all samples were in accordance to the legal Egyptian standards. Similar results were reported by **Tarakçı and Küçüköner (2003b)**.

### Sensory Evaluation of Flavoured Probiotic Stirred Yoghurt

Scores of organoleptic properties of control and flavoured probiotic stirred yoghurts are shown in Table 8. Stirred yoghurt samples blended with 10% mango pulp scored the highest points 41.50 (M<sub>2</sub>) for the flavour whereas lowest score was 38.60 (M<sub>1</sub>). In case of body and texture, the highest score was recorded for treatment combination  $A_3$  (32.70) while the score decreased on further addition of mango pulp and the lowest score was recorded in  $M_1$ (30.50). Concerning appearance, the highest score was 9.30 (A<sub>2</sub>) and the lowest score was 7.80 (M<sub>1</sub>). The scores for overall acceptability recorded highest score for the treatment A<sub>2</sub> (91.40) over all the other treatment combinations. It is observed that the score for all the parameters increased with increase in apple and mango pulps proportion in stirred yoghurt to 10%, thereafter it decreased proportionately. The values for all parameters varied. The general trend of these results are in agreement with those reported by **Raut** *et al.* (2015).

#### Conclusion

Addition of apple and mango pulps to probiotic flavoured stirred yoghurt presents a potential prebiotic effect, increased the nutritional quality, and improved sensory attributes, rheological, microbiological and physicochemical properties of voghurt. Among fruit flavoured voghurt ones, 10% apple flavoured yoghurt scored the highest rank as compared to others fruit flavoured yoghurt and plain yoghurt. The evidence from this study suggests that addition of apple and mango pulps increased acceptability of probiotic flavoured stirred yoghurt, greatly activated bifidobacterial and lactobacilli bacterial growth and activity. The middle concentration levels were superior over higher concentration levels of apple and mango pulps fortification.

Yoghurt sample		s count 10²/g)	Mould (cfu×	s count 10 <sup>1</sup> /g)	Coliforms count (cfu×10 <sup>1</sup> /g) Storage period (day)			
-		e period ay)	Storage (da					
	0	15	0	15	0	15		
С	30	38	ND	2	ND	4		
$\mathbf{A}_{1}$	41	48	ND	5	ND	6		
$A_2$	44	53	ND	7	ND	7		
$A_3$	50	61	ND	11	ND	9		
$\mathbf{M}_{1}$	32	40	ND	3	ND	5		
$\mathbf{M}_{2}$	35	42	ND	6	ND	7		
$M_3$	39 44		ND	9	ND	8		

Table 7. Yeasts, moulds and coliforms counts of flavoured probiotic stirred yoghurt during storage period at 5°C for 15 days

ND: Not Detected - For Note see: Table 2.

Table 8. Sensory evaluation of flavoured probiotic stirred yoghurt during storage period at 5°C for 15 days

Yoghurt sample	0 ()		Bo	Body and texture (35)			Ар	peara	ance (	10)		Acidity (10)				Overall acceptability (100)				
Storage period (day)			Storage period (day)			Storage period (day)			Storage period (day)				Storage period (day)							
	0	5	10	15	0	5	10	15	0	5	10	15	0	5	10	15	0	5	10	15
С	40.1	40.0	39.1	35.0	30.1	29.0	28.0	27.2	8.3	8.3	8.0	7.5	8.3	8.0	7.0	7.0	86.8	84.5	82.0	80.5
$\mathbf{A}_{1}$	40.4	40.4	39.5	35.5	31.2	30.3	29.3	28.0	7.85	7.5	7.5	7.0	8.55	8.0	7.0	7.0	88.0	87.0	85.5	83.0
A <sub>2</sub>	41.2	41.0	39.5	38.8	32.2	31.6	30.5	29.6	9.3	8.5	8.5	8.0	8.7	8.5	7.5	7.0	91.4	91.0	90.5	88.0
A <sub>3</sub>	41.0	41.0	39.0	37.5	32.7	30.9	29.8	28.5	8.1	8.0	8.0	7.5	8.7	8.0	7.0	6.0	90.5	88.0	86.5	85.0
$M_1$	38.6	38.5	37.0	34.8	30.5	29.4	28.3	27.5	7.8	7.5	7.3	7.0	8.1	7.5	7.5	7.0	85.0	82.5	81.0	78.5
$M_2$	41.5	40.7	40.0	38.3	31.5	30.8	29.5	28.7	8.3	8.0	8.0	7.7	8.0	7.5	7.0	6.0	89.3	87.5	85.5	83.0
$M_3$	40.5	39.0	37.7	37.0	31.4	30.0	29.0	28.0	8.1	7.8	7.5	7.5	8.0	7.0	6.0	6.0	88.0	86.0	84.5	81.0

For Note see: Table 2.

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## تدعيم اليوغورت الحيوى المُقلب بإضافة لُب التفاح ولُب المانجو

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تم استخدام أب التفاح والمانجو في تدعيم اليو غورت الحيوي المُقلب المنكَّه، تم تصنيع اليو غورت الحيوي المقلب المنكَّه من الحليب البُقري المدعَّم بلُب التفاح والمانجو عند مستويات ، ، ، ، ، و `١٠،٥ و `١٠% وتم تخزين اليوغورت الناتج من جميع المعاملات على درجة حرارة ٥ م لمدة ١٥ يوماً لدراسة الخواص الفيزيائية والكيميائية والميكروبيولوجية والحسية والخواص اللونية (L\*, a\*, b\*) لعينات اليوغورت خلال فترة التخزين ، أدى إضافة لب التفاح والمانجو للحليب البقرى لزيادة الحموضة، الجوامد الصلبة الكلية ولكن حدث انخفاض في محتوى البروتين الكلي والدهن الكلي وقيم الرقم الهيدروجيني لليوغورت الحيوي المُقلب المنكَّه. أيضاً أدى إضافة لب التفاح والمانجو لتحسين نِشاط البادئ، وزيادة العدد الكلى للميكروبات والميكروبات الحيوية والعصويات اللبنية في اليوغورت الحيوي المُقلب المُنكَّه، وقد تفاوتت الخصائص اللونية (L\*, a\*, b) وفقاً لأنواع لُب الفاكهة المُستخدمة في اليوغورت والتي تسببت في حدوث تغيرات في لون عينات اليوغورك وارتبطت هذه الملاحظات بمستوى إضافة أب التفاح والمانجو. ونتيجةً للتقييمات الحسية حصلت عينة اليوغورت المحتوبة على ١٠% من لُب التفاح على أفضل درجات القبول العام. ويمكن التوصية باستخدام لُب الفاكهة بنسبة • ١% وخصوصاً لُب التفاح لإنتاج اليو غورت الحيوي المُطعَّم بالفواكه على نطاق و أسع.

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