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GROWTH AND SURVIVAL OF PROBIOTIC BACTERIA IN FERMENTED FLAVOURED SOY MILK DRINKS DURING STORAGE

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ABSTRACT: This study investigated the viability of probiotic bacteria and changes in pH in fermented flavoured soy milk drinks using ABY-1 starter culture which contains *Streptococcus thermophilus*, *Lactobacillus delbrueckii* subsp. *bulgaricus*, *L. acidophilus* LA-5 and Bifidobacterium BB-12. Soy milk supported the growth of all tested organisms through 21 days of storage at 5°C. Fermented soy milk was mixed with 4% sucrose and 15% of 3 kinds of fruits, banana, guava, and mango to produce ferminted flavoured soy milk drinks. All formulations showed probiotic viabilities ranging from 5 to 9 log cfu/g, and fruit pulps did not affect the probiotic viabilities.

Key words: Soy milk, soy milk drinks, probiotic bacteria.

INTRODUCTION

Probiotics are defined as live microorganisms administered in adequate amounts confer a health benefit on the host **(FAO/WHO, 2001)**. Probiotics reported to give a several beneficial health advantages, they help maintaining the intestinal flora composition and balance, and raising the resistance to pathogens. There is a lot of efforts to produce alternative healthy products of cow milk provides and achieve the market challenge. Recently there is increasing demands on fermented milk products as a result of the increasing attentions of consumers for its impact on health, so seeking of probiotic functional foods is growing faster. Bifidobacteria are often incorporated in fermented dairy products to increase their therapeutic value **(Driessen and De Boer, 1989; Holcomb *et al.*, 1991; Ishibashi and Shimamura, 1993; Dinakar and Mistry, 1994; Blanchette and Roy, 1995; Samona *et al.*, 1996)**.

Soybean is incorporated in consumers diet due to its nutritional characteristics such as dietary fibers, poly-unsaturated fatty acids, high quality proteins, besides its role of reducing cardiovascular diseases, type 2 diabetes, cancer, and osteoporosis **(Xiao, 2008; Chen *et al.*, 2010; Rinaldoni *et al.*, 2012).**

The undesirable beany favour of the soy milk limits its consumption beside it's oligosaccharides contents (stachyose and raffinose) that leads to intense discomfort **(Yeo and Liong, 2010)**.

The probiotic microorganisms most widely used are strains belonging to the lactobacillus and bifidobacteria genera **(Saxelin *et. al.*, 2005)**.

Soy milk fermentation, especially when using lactic acid bacteria, for producing soy yoghurt, may improve its flavour and texture, as well as enhance its nutritonal and beneficial health properties **(Donkor *et al.*, 2005; Cruz *et al.*, 2009)**.

Yoghurt drink gained popularity in Egypt. So soy yoghurt may achieve the same popularity if it find the suitable awareness ways to the consumer. It is familiar to incorporate Bifidobacteria in yoghurt and soy milk. So, this investigation aimed to evaluate the growth of bifidobacteria and lactic acid bacteria, and to measure the acid production during the fermentation of soy milk, and its viability in soy milk fermented drinks during subsequent storage of the drinks at 5°C for 21 days.

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MATERIALS AND METHODS

Materials

Buffalo's whole fresh milk (5.5% fat, 4.3% protein, 0.81% ash and 5% lactose) was obtained from Dairy Technology Unit, Food Science Department, Faculty of Agriculture, Zagazig University.

Soybeans (yellow variety) were obtained from Botany Dept., Faculty of Agriculture, Cairo University, Egypt.

Cane sugar, and fresh fruit (guava, mango and banana) were purchased from the local market at Zagazig, Sharkia Governorate, Egypt.

Stabilizers [Guar gum E412, sodium carboxymethyl cellulose E466 and mono and diglyceride of fatty acid E471 (1:1:1)], were obtained from the Egyptian Company for Dairy Products and Food Additives "EGY- DAIRY" (10^{th} of Ramadan city, Sharkia Governorate, Egypt).

Freeze dried DVS (nutrish ABY-1) yoghurt cultures containing *Streptococcus thermophilus*, *Lactobacillus delbrueckii* subsp. *bulgaricus*, *L. acidophilus* LA-5 and Bifidobacterium BB-12 were obtained from Chr. Hansen Inc. Laboratories, Denmark, by Misr Food Additives (MIFAD), Egypt.

Methods

Preparation of soy milk

Soy milk was prepared in the laboratory from whole soybeans using the procedure described by **Bourne *et al.* (1976)**.

Preparation of fermented soy milk

Stabilizers were added to the soy milk at level of 0.5%, and then homogenized at 60ºC, 400Kpa. The soy milk was heated at 85°C for 10 min, cooled to 42°C and inoculated with freeze dried ABY-1 culture (3%), distributed in 200 ml sterile plastic containers followed by incubation at 42°C for 12 hours. The set curd was refrigerated for 3 hours and used for flavoured soy drink preparation.

Preparation of fruit pulp and fruit fermented flavoured soy milk drinks

Fruit pulp was prepared by washing, the fruits of guava, mango and banana, then cleaned thoroughly with water. The fruits were peeled and cut into pieces, the cut pieces were ground in a blender then heat treated at (63^{o}C for 30 min).

The fruit pulp (guava or mango or banana) of each was added at ratio 15% to the fermented soy milk and mixed gently. SY (control soy yoghurt without any additives), SYS (soy yoghurt and 4% sucrose without the fruit pulp), SYSB (with Banana and 4% sucrose), SYSG (with guava pulp and 4% sucrose), SYSM (with mango pulp and 4% sucrose), and BY (buffalo yoghurt) as a general control. Result soy milk fermented drinks were stored at 5^{o}C for 21 days.

Methods of Analyses

Bacteriological examination

Fermented flavoured soy milk drinks treatments were subjected to bacteriological examination when fresh, then after 7, 14 and 21 days of storage at 5^{o}C. All microorganisms which inoculated into fermented soy milk treatments were enumerated by using differential media and methods.

Serial delutions of flavoured soy milk fermented drinks were made. The results were expressed as log colony-forming units per gram (log cfu/g) of sample and the viability of each culture in different treatments was calculated according to **Paseephol and Sherkat (2009)**.

Streptococcus salivarius spp. *thermophilus* count

M17 agar (Difco Laboratories) was used to enumerate streptococci in fermented flavoured soy milk drinks treatments **(Dave and Shah, 1996)**. Plates were incubated in aerobic incubator at 37°C for 72 hr.

Lactobacillus delbrueckii spp. *bulgaricus* count.

Acidified MRS (pH 5.2) agar (Difco Laboratories) was used for enumeration **(Dave and Shah, 1997)**. Plates were incubated under anaerobic conditions at 37° C for 72 hr.

Lactobacillus acidophilus count.

MRS agar (Difco Laboratories, Detroit, MI) with 0.20% oxgall (Difco Laboratories) was used **(Marshall, 1992)**. Plates were incubated aerobically at 37°C for 72 hr.

***Bifidobacterium bifidum* count**

MRS agar with added neomycin-paromomycin- nalidixic acid- lithium chloride (NPNL) solution was used to enumerate *B.bifidum* **(Martin and Chou, 1992)**. Bacteria were grown in a fresh medium under anaerobic conditions at 37°C for 72 hr., **(Laroia and Martin, 1991)**. The solution of NPNL broth with 1% L–cysteine was prepared according to **Karagu`l-Yuceer *et al.* (2001)**.

Titratable acidity and pH

Titratable acidity and pH were determined as given by **AOAC (1995)**. The results of titratable acidity were recorded as percentage of lactic acid. The pH of the various treatments was determined using a pH-meter (model Horiba, B-211, Shimadzu Analytical Instruments, Kyoto, Japan). All measurements were carried out in triplicates.

Statistical Analysis

Statistical analysis of the obtained data was carried out according to the methods described by **Clarke and Kempson (1997)**. Experiments were repeated in triplicates and each analysis was carried out in duplicates and the average of results were tabulated.

RESULTS AND DISCUSSION

Titratable Acidity

Table 1 shows the titratable acidity values of fermented flavoured soy milk drinks treatments during refrigerated storage.

The changes in titratable acidity occurred to a greater or lesser degree, depending on the chemical composition of the product especially the viable fermentable sugars, the cold storage temperature, and time. Moreover, protein content can influence the acidity of dairy products, as proteins act as a buffer due to the large number of groups that can reversibly interact with protons **(Pimentel *et al.*, 2012).** Higher product acidity can protect the product from the development of spoilage microorganisms, which increases shelf life and it does not change the product's sensory or technological characteristics **(Pimentel *et al.*, 2015)**.

pH Values

The pH values of fermented flavoured soy milk drinks treatments during refrigerated storage period are shown in Table 2. The initial pH values for the fresh different soy milk drink types ranged from 4.81 to 5.11. The pH of all treatments decreased slightly during storage and did not drop under 4.33 at the end of storage. The drop in the pH was almost similar for all of the treatments, and between 0.07 and 0.22 pH units throughout storage.

There was no major difference in pH values or relative drop in pH values at 5^{o}C to 21 days of storage. It is unlikely that these changes would affect viability.

Dave and Shah (1997) obtained pH of 4.16 and 4.40 after 35 days of storage (5°C) in probiotic yogurts, when the initial pH values were 4.33 and 4.61, respectively. **Gilliland *et al.* (2002)** obtained pH values of 4.1 and 4.2 at the end of 35 days of refrigerated (5°C) storage of yogurt type products fermented with *S. thermophilus, L. acidophilus*, bifidobacteria or *L. casei*; their initial pH values were 4.7 and 4.8, respectively. **Gueimonde *et al.* (2004)** analyzed 14 commercial fermented milks and observed pH values around 3.9 to 4.2. Results of the present work are similar to these reports, corroborating the residual acidification during storage.

Donkor *et al.* (2007) observed production of acetic and lactic acids in milk fermented by *L. acidophilus* and *L. casei* associated with yogurt culture during 28 days of cold storage. **Korbekandi *et al.* (2008)** reported similar results in yogurts with *L. casei*. **Bedani *et al.* (2014)** didn't find significant difference ($p>0.05$) between soy yoghurt formulations in each storage period evaluated and the addition of fruit pulps and essences did not influence the pH values of soy yoghurt during refrigerated storage. But some studies have shown that the incorporation of fruit pulps and juices may reduce the pH values in soy-based products **(Granato *et al.*, 2010; Osundahunsi *et al.*, 2007)**.

Cell Viability

Streptococcus salivarius* spp. *thermophilus

Table 3 shows the total viable counts of *Streptococcus salivarius* spp. *thermophilus*, of

Table 1. The changes in titratable acidity (as a percentage of lactic acid) of fermented flavoured soy milk drinks during storage period at 5°C for 21 days

Storage period (day)	Treatment						Mean effect
	BY	SY	SYS	SYSG	SYSM	SYSB	
Fresh	0.65b	0.43d	0.53c	0.75a	0.72a	0.8a	0.64±0.13D
7	0.82c	0.56d	0.64d	0.93ab	0.86bc	0.97a	0.79±0.15C
14	0.93b	0.64d	0.75c	0.98b	0.93b	1.13a	0.89±0.17B
21	1.01b	0.77c	0.93bc	1.15ab	1.05ab	1.24a	1.02±0.17A
Mean effect	0.85±0.14C	0.60±0.13E	0.71±0.15D	0.95±0.15B	0.89±0.13C	1.03±0.18A	

Mean (±SE). Values with small letters in the same row and values with capital letters in the column or row having different superscripts differ significantly ($p \leq 0.05$). BY (Buffalo beverage), SY (control soy milk fermented drinks without any additives), SYS (soy milk fermented drinks and 4% sucrose without the fruit pulp), SYSB (soy milk fermented drinks with banana and 4% sucrose), SYSG (soy milk fermented drinks with guava pulp and 4% sucrose), and SYSM (soy milk fermented drinks with mango pulp and 4% sucrose).

Table 2. The changes in pH value of fermented flavoured soy milk drinks during storage period at 5°C for 21 days

Storage period (day)	Treatment						Mean effect
	BY	SY	SYS	SYSG	SYSM	SYSB	
Fresh	4.87bc	5.11a	4.92b	4.82c	4.84c	4.81c	4.89±0.10A
7	4.76bc	4.94a	4.81b	4.73bc	4.75bc	4.68c	4.77±0.09B
14	4.67bc	4.85a	4.74b	4.55d	4.62cd	4.46e	4.64±0.13C
21	4.56c	4.77a	4.63b	4.41d	4.46d	4.33e	4.52±0.15D
Mean effect	4.71±0.12C	4.91±0.13A	4.77±0.11B	4.62±0.16E	4.66±0.15D	4.57±0.19F	

Mean (±SE). Values with small letters in the same row and values with capital letters in the column or row having different superscripts differ significantly ($p \leq 0.05$). BY (Buffalo beverage), SY (control soy milk fermented drinks without any additives), SYS (soy milk fermented drinks and 4% sucrose without the fruit pulp), SYSB (soy milk fermented drinks with banana and 4% sucrose), SYSG (soy milk fermented drinks with guava pulp and 4% sucrose), and SYSM (soy milk fermented drinks with mango pulp and 4% sucrose).

Table 3. Total viable counts of *Streptococcus salivarius* spp. *thermophilus*, of fermented flavoured soy milk drinks during storage period at 5°C for 21 days (log cfu/g)

Storage period (day)	Treatment						Mean effect
	BY	SY	SYS	SYSG	SYSM	SYSB	
Fresh	8.09a	6.32a	7.56a	8.16a	8.04a	8.23a	7.73±0.91A
7	8.15a	6.43a	7.85a	8.26a	8.13a	8.51a	7.88±1.09A
14	8.02a	5.77a	7.43a	8.14a	8.05a	8.31a	7.62±1.27A
21	7.46a	5.41b	6.58ab	7.71a	7.46a	7.88a	7.08±1.11A
Viability (%)	92.21	85.60	87.04	94.49	92.79	95.75	
Mean Effect	7.93±0.77A	5.98±0.79B	7.35±0.91A	8.06±0.080A	7.92±0.94A	8.23±0.88A	

Mean (±SE). Values with small letters in the same row and values with capital letters in the column or row having different superscripts differ, significantly ($p \leq 0.05$). BY (Buffalo beverage), SY (control soy milk fermented drinks without any additives), SYS (soy milk fermented drinks and 4% sucrose without the fruit pulp), SYSB (soy milk fermented drinks with banana and 4% sucrose), SYSG (soy milk fermented drinks with guava pulp and 4% sucrose), and SYSM (soy milk fermented drinks with mango pulp and 4% sucrose).

fermented flavoured soy milk drinks during storage period. The SYSB treatment has the highest initial and final cell count, 8.23 and 7.88 log cfu/g respectively, among all of them during the 21 days of storage. SYSG followed it, then SYSM, BY, SYS then the control.

The growth curve of the BY, and SYSM was similar up to the end of storage period. All treatments counts were increased gradually at the 7th day of storage then decreased up to the 21th day. The SYS treatment has the fastest decrease rate.

At the end of storage period, the highest final viable cell count was 7.88 and the lowest was 5.41 log cfu/g which occurred in SYSB and SY respectively. After 21 days of storage, *Streptococcus salivarius* spp. *thermophilus* exhibited the highest final viable cell counts and *Bifidobacterium bifidum* has the lowest which occurred in SYSB and SY, respectively.

Lactobacillus delbrueckii* spp. *bulgaricus

Table 4 shows the viable counts of *Lactobacillus delbrueckii* spp. *bulgaricus* during 21 days of refrigerated storage. At the 7th day SYSB reached its maximum population among treatments 8.18 log cfu/g followed by SYSG, SYSM, BY, SYS then the control. The initial counts of SYS was lower than the other treatments but the growth curve between the 7th day and the 14th was similar to the SYSM then falled down at the end of the storage period.

All treatments counts of *Lactobacillus delbrueckii* spp. *bulgaricus* were increased gradually up to the 7th day of storage then they decreased up to the 21th day. The SY treatment has the fastest decrease rate. After 21 days of storage, SYSB has the highest final viable number 7.1 log cfu/g, and SY has the lowest final viable number 4.68 log cfu/g.

The highest viable counts at the end of storage period was in each of SYSB, SYSG, SYSM, BY, SYS, and control treatments, respectively.

Lactobacillus delbrueckii spp. *bulgaricus* grew in soy milk supplemented with prebiotics, with viable counts ranging from 4.68 to 8.18 log cfu/g.

According to **Dave and Shah (1996),** hydrogen peroxide produced by *L. delbruecki* ssp. *bulgaricus* bacteria is the most important viability-reducing factor during refrigerated storage.

Table 4. Total viable counts of *Lactobacillus delbrueckii* spp. *bulgaricus*, of fermented flavoured soy milk drinks during storage period at 5°C for 21 days (log cfu/g)

Storage period (day)	Treatment						Mean effect
	BY	SY	SYS	SYSG	SYSM	SYSB	
Fresh	7.83a	6.01a	7.34a	8.02a	7.93a	8.02a	7.52±0.96A
7	7.91a	6.24a	7.77a	8.11a	7.79a	8.18a	7.66±1.05A
14	7.64a	5.55b	7.36a	7.65a	7.46a	7.71a	7.22±1.01A
21	6.51a	4.68b	5.89a	6.86a	6.57a	7.1a	6.26±0.97B
Viability (%)	83.14	77.87	80.25	85.54	82.85	88.53	
Mean Effect	7.47±0.80A	5.62±0.86B	7.09±0.98A	7.66±0.83A	7.43±1.00A	7.75±0.80A	

Mean (±SE). Values with small letters in the same row and values with capital letters in the column or row having different superscripts differ significantly ($p \leq 0.05$). BY (Buffalo beverage), SY (control soy milk fermented drinks without any additives), SYS (soy milk fermented drinks and 4% sucrose without the fruit pulp), SYSB (soy milk fermented drinks with banana and 4% sucrose), SYSG (soy milk fermented drinks with guava pulp and 4% sucrose), and SYSM (soy milk fermented drinks with mango pulp and 4% sucrose).

Lactobacillus acidophilus

The viability of *L. acidophilus* in fermented flavoured soy milk drink formulations throughout storage is shown in Table 5.

The highest number among all of the treatments at the end of storage period was in SYSB which was 7.58 log cfu/g, followed by SYSG, SYSM, BY, SYS then the control in order.

All treatments counts were increased gradually up to the 7th day of storage then they decreased up to the 21th day.

The initial counts and growth curve of the SYSB and SYSG was similar until the 7th day then they exchange the growth rates between them to the 21 days which the SYSB has the highest viable counts. The SYS treatment has the fastest decrease rate. All populations increased at the 7th day of storage then decreased at the end of storage period.

Ranadheera *et al.* (2012) reported that the populations of probiotic bacteria in plain and stirred fruit yoghurts made from goat's milk decreased in all formulations during 4 weeks of storage and the higher loss in cell viability was observed for *L. acidophilus* La-5 than for bifidobacteria and propionibacteria, but the addition of commercial fruit appeared to support the viability of *L. acidophilus* La-5, with higher counts in fruit yoghurts than in plain yoghurt throughout storage.

Bedani *et al.* (2014) found that *L. acidophilus* and *Bifidobacterium animalis* populations remained above 8 log cfu /g between the first and the 28th day of storage in the different soy fruit products. They also reported that addition of pulps and essences in SY did not have influence upon *L. acidophilus* La-5 and *B. animalis* Bb-12 viability during the storage. Even though certain variations in the *L. acidophilus* and *B. animalis* populations were observed among SY formulations in each storage period evaluated. These changes are of little microbiological significance, since they are always below 0.5 log cfu/g.

Kailasapathy *et al.* (2008) verified that the addition of either 5 or 10 g/100 g of fruit preparations to yoghurts had no significant effect on the viability of *L. acidophilus* LAFTI L10 and LAFTI B94 during 35 days of storage. **Oliveira *et al.* (2006)** found that *Lactobacillus acidoiphilus* counts decreased during cold storage until 28 days to a level that doesn't fulfill the minimum viable counts to reach health beneficial effects. **Wang *et al.* (2002)** found that with or without adding sucrose, no marked changes in the counts of *B. infantis*, *B. longum* or *L. acidophilus* were observed during 10 days of storage at 5°C.

Table 5. Total viable counts of *Lactobacillus acidophilus* of fermented flavoured soy milk drinks during storage period at 5°C for 21 days (log cfu/g)

Storage period (day)	Treatment						Mean effect
	BY	SY	SYS	SYSG	SYSM	SYSB	
Fresh	7.54a	6.44a	7.34a	8.15a	8.02a	8.12a	7.60±0.91A
7	7.91ab	6.57b	7.71ab	8.83a	8.11ab	8.87a	8.00±0.99A
14	7.76a	6.12a	7.42a	8.44a	7.73a	8.19a	7.61±1.04A
21	6.79a	5.13b	6.14ab	7.36a	7.23a	7.58a	6.70±1.04B
Viability (%)	90.05	79.66	83.65	90.31	90.15	93.35	
Mean effect	7.50±0.77AB	6.06±0.82C	7.15±0.85B	8.19±0.97A	7.77±0.76AB	8.19±0.79A	

Mean (±SE). Values with small letters in the same row and values with capital letters in the column or row having different superscripts differ significantly ($p \leq 0.05$). BY (Buffalo beverage), SY (control soy milk fermented drinks without any additives), SYS (soy milk fermented drinks and 4% sucrose without the fruit pulp), SYSB (soy milk fermented drinks with banana and 4% sucrose), SYSG (soy milk fermented drinks with guava pulp and 4% sucrose), and SYSM (soy milk fermented drinks with mango pulp and 4% sucrose).

Buriti *et al.* (2007) reported that the *L. acidophilus* La-5 viability decreased approximately 5.0 log cfu/g in mousses containing passion fruit pulp during 21 days of refrigerated storage, while this probiotic strain population remained above 6.0 log cfu/g in mousses with guava pulps until the end of a storage period of 21 days.

Oliveira *et al.* (2006) found that *Bifidobacterium lactis* and bacterial culture remained stable during cold storage until 28 days. **Matsuyama *et al.* (1992)** observed higher counts of *B. longum* in soy milk fermented simultaneously with *L. acidophilus* than in soy milk fermented by *B. longum* alone. However, **Wang *et al.* (2002)** recorded that, at the end of fermentation, the final counts of *B. longum* in the mixed culture with lactic acid bacteria were significantly less ($P<0.05$) than the final counts in the pure culture.

Bifidobacterium bifidum

Changes in the viable counts of *Bifidobacterium bifidum* during storage period are shown in Table 6 *B. bifidum* population in soy yoghurt treatments increased at the 7th day of storage then decreased slightly in the formulations throughout storage, but the rapid lose happened in SYS treatment after that.

Wang *et al.* (2002) found that the growth of *B. longum* in the mixed cultures with lactic acid bacteria was generally similar to growth of the organism alone, and after 24 hr., of incubation, the counts of *B. longum* in the mixed culture with lactic acid bacteria were significantly higher ($P<0.05$) than the counts in a pure culture.

In general, the increasing of the storage temperature increases the metabolic activities of bacterial cells, thereby causing an increase in their death rate, so storing yogurt at 8°C than 5°C for 10 days may resulted in the lower viability of *L. Acidophilus* compared to that stored at 5°C **(Mortazavian *et al.*, 2007).**

probiotic micro-organisms often show poor viability in commercial preparations, and several factors have been identified in fermented milk that can affect their viability, such as the pH and acidity levels, presence of other micro-organisms, temperature of incubation and/or the presence of oxygen **(Shah *et al.*, 1995; Kailasapathy and Rybka, 1997; Shah, 2000).**

Kamaly (1997) found that the growth of probiotics was not affected by supplementation with carbohydrates such as lactose, glucose and galactose and protein hydrolysates such as yeast extract, peptone and casitone in soy milk. However, such supplementation significantly affected the production of acids.

Table 6. Total viable counts of *Bifidobacterium bifidum* of fermented flavoured soy milk drinks during storage period at 5°C for 21 days (log cfu/g)

Storage period (day)	Treatment						Mean effect
	BY	SY	SYS	SYSG	SYSM	SYSB	
Fresh	7.48ab	6.02b	6.61ab	8.11a	8.07a	8.14a	7.40±1.04A
7	7.39a	6.27a	7.03a	8.32a	8.27a	8.55a	7.63±1.19A
14	7.04a	5.91a	6.51a	8.01a	7.92a	8.03a	7.23±1.10A
21	6.78a	5.01b	5.72ab	7.51a	7.32a	7.62a	6.66±1.19B
Viability (%)	90.64	83.22	86.54	92.60	90.71	93.61	
Mean effect	7.17±0.85A	5.80±0.83B	6.46±0.80B	7.98±0.82A	7.89±0.91A	8.08±0.69A	

Mean (±SE). Values with small letters in the same row and values with capital letters in the column or row having different superscripts differ significantly ($p \leq 0.05$). BY (Buffalo beverage), SY (control soy milk fermented drinks without any additives), SYS (soy milk fermented drinks and 4% sucrose without the fruit pulp), SYSB (soy milk fermented drinks with banana and 4% sucrose), SYSG (soy milk fermented drinks with guava pulp and 4% sucrose), and SYSM (soy milk fermented drinks with mango pulp and 4% sucrose).

Fruit pulps employed in the present study did not contain any added preservative that might cause the loss of viability, since they were natural, and pasteurised pulps.

Sensory Evaluation

The organoleptic properties of the fermented flavourd soy milk drinks are shown in Table 7. Panel of seven judges, familiar with fermented milks were chosen from the staff members of the Faculty of Agriculture, Zagazig University, according to the scheme described by **Farag *et al.* (2007)**.

Results indicated that fermented flavoured soy milk drinks showed higher scores than fermented soy milk without fruit pulp at the fresh stage. Moreover, SYSM showed the highest score. Total scores were decreased for all treatments up to the end of storage period. There was a trend for higher acceptability scores of soy milk fermented drinks falvoured with mango pulp throughout 14 days of storage. On day 21, SYSM presented significantly higher acceptability scores, compared to SY, SYS, SYSB and SYSG. Similarly, **Kumar and Mishra (2003)** observed that the overall acceptability of mango soy fortified yoghurt formulation increased with the proportion of mango pulp. The improved flavour to the fruit pulp has a masking effect against the beany soy milk flavour. A possible explanation for the relatively low acceptability scores obtained in the present study may be related to the volunteers' lack of habit to consume soy-based products, particularly fermented soy products.

Conclusions

This study showed that fermented flavoured soy milk drinks supported the viablilty of Lactobacillus and Bifidobacterium and the other microorganisms in the ABY-1 starter culture. The viabilities ranged from 5 to 9 log cfu/g during the 21 days of storage at 5°C, and the addition of fruit pulps did not affect the viability of the probiotic bacteria. Acceptability was higher for mango fermented soy milk drink, this difference was significant upon the 21 days of storage. Somehow there was acceptability improvment of SY through the addition of tropical fruits pulps.

Table 7. Organoleptic properties of fruit soy fermented beverage during storage period

Property	Storage period (day)	Treatment						Mean effect
		BY	SY	SYS	SYSG	SYSM	SYSB	
Flavour (45)	Fresh	38.50^{b}	31.24^{d}	36.40^{c}	41.25^{a}	40.50^{a}	39.50^{ab}	37.89 ± 3.68^{A}
	7	35.71^{bc}	27.56^{d}	34.12^{c}	38.25^{a}	36.41^{ab}	38.50^{a}	35.09 ± 4.03^{B}
	14	30.25^{b}	25.32^{d}	28.22^{c}	36.25^{a}	35.25^{a}	34.58^{a}	31.64 ± 4.29^{C}
	21	28.70^{b}	22.18^{c}	26.41^{b}	33.78^{a}	23.71^{c}	31.45^{a}	27.70 ± 4.64^{D}
Mean effect		33.29 ± 4.36^{BC}	26.57 ± 3.58^{D}	31.28 ± 4.51^{C}	37.38 ± 3.27^{A}	33.96 ± 6.56^{BC}	36.00 ± 5.63^{AB}	
Consistency (35)	Fresh	30.14^{b}	26.12^{c}	28.25^{bc}	33.40^{a}	34.53^{a}	32.51^{a}	30.82 ± 3.56^{A}
	7	25.50^{b}	22.15^{c}	23.45^{c}	32.34^{a}	32.50^{a}	31.50^{a}	27.90 ± 4.69^{B}
	14	23.50^{c}	19.35^{e}	21.74^{d}	30.51^{a}	31.50^{a}	28.50^{b}	25.85 ± 4.83^{C}
	21	20.35^{c}	17.46^{d}	19.54^{c}	29.25^{a}	30.15^{a}	26.25^{b}	23.83 ± 5.26^{D}
Mean effect		24.87 ± 4.16^{C}	21.27 ± 3.75^{D}	23.24 ± 3.41^{C}	31.37 ± 2.11^{AB}	32.17 ± 2.11^{A}	29.69 ± 3.39^{B}	
Acidity (10)	Fresh	7.48^{ab}	6.86^{b}	7.22^{ab}	7.69^{ab}	8.01^{a}	7.55^{ab}	7.46 ± 0.63^{C}
	7	7.54^{bc}	7.11^{c}	7.43^{bc}	7.85^{ab}	8.26^{a}	7.65^{bc}	7.64 ± 0.54^{C}
	14	7.78^{bc}	7.37^{c}	7.58^{c}	8.51^{a}	8.33^{ab}	8.22^{ab}	7.96 ± 0.60^{B}
	21	8.22^{bc}	7.54^{c}	7.82^{bc}	8.57^{ab}	9.13^{a}	8.41^{b}	8.28 ± 0.72^{A}
Mean effect		7.75 ± 0.68^{CD}	7.22 ± 0.55^{E}	7.51 ± 0.49^{DE}	8.15 ± 0.58^{AB}	8.43 ± 0.61^{A}	7.95 ± 0.51^{BC}	
Appearance (10)	Fresh	8.5^{b}	7.14^{d}	7.63^{c}	8.85^{ab}	9.24^{a}	9.14^{a}	8.41 ± 0.85^{A}
	7	8.58^{b}	7.37^{d}	7.88^{c}	8.61^{b}	9.54^{a}	9.25^{a}	8.53 ± 0.81^{A}
	14	8.26^{b}	6.23^{d}	7.33^{c}	8.55^{b}	9.21^{a}	9.01^{a}	8.09 ± 1.10^{B}
	21	7.28^{b}	6.11^{c}	7.03^{b}	7.41^{b}	8.5^{a}	8.33^{a}	7.44 ± 0.91^{C}
Mean effect		8.15 ± 0.71^{B}	6.71 ± 0.70^{D}	7.46 ± 0.48^{C}	8.35 ± 0.65^{B}	9.12 ± 0.46^{A}	8.93 ± 0.46^{A}	
Total scores (100)	Fresh	84.62^{b}	71.36^{d}	79.50^{c}	91.19^{a}	92.28^{a}	88.70^{a}	84.60 ± 7.83^{A}
	7	77.33^{b}	64.19^{d}	72.88^{c}	87.05^{a}	86.71^{a}	86.90^{a}	79.17 ± 8.91^{B}
	14	69.79^{c}	58.27^{e}	64.87^{d}	83.82^{a}	84.29^{a}	80.31^{b}	73.55 ± 10.19^{C}
	21	64.55^{d}	53.29^{f}	60.80^{e}	79.01^{a}	71.49^{c}	74.44^{b}	67.26 ± 9.03^{D}
Mean effect		74.07 ± 8.06^{B}	61.77 ± 7.28^{D}	69.51 ± 7.73^{C}	85.26 ± 4.99^{A}	83.69 ± 7.91^{A}	82.58 ± 6.05^{A}	

Mean (±SE). Values with small letters in the same row and values with capital letters in the column or row having different superscripts differ significantly ($p \leq 0.05$). BY (Buffalo beverage), SY (control soy milk fermented drinks without any additives), SYS (soy milk fermented drinks and 4% sucrose without the fruit pulp), SYSB (soy milk fermented drinks with banana and 4% sucrose), SYSG (soy milk fermented drinks with guava pulp and 4% sucrose), and SYSM (soy milk fermented drinks with mango pulp and 4% sucrose).

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نمو وبقاء بكتيريا البروبيوتك فى مشروبات لبن فول الصويا المتخمرة المنكهة خلال التخزين

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أجريت هذه الدراسة لتقييم نمو بكتيريا البروبيوتيك والتغيرات في درجة الحموضة في المشروبات المتخمره المصنوعه من لبن الصويا باستخدام بادىء ABY-1 واللذي يحتوي على بكتيريا *Streptococcus thermophilus*, *Lactobacillus delbrueckii* subsp. *bulgaricus*, *L. acidophilus* LA-5 and Bifidobacterium BB-12، وقد دعم لبن الصويا المتخمر نمو تلك البكتيريا خلال ٢١ يوما من التخزين على درجة حرارة ٥°م، حيث تم خلط لبن الصويا المتخمر مع ٣ أنواع من الفواكه مثل الموز والجوافة والمانجو بنسبة ١٥% لإنتاج مشروبات لبن الصويا المتخمره المنكهه، وأظهرت جميع المعاملات إمكانية دعم نمو تلك البكتيريا حيث تراوحت أعداد البكتيريا الحية فى المنتجات ما بين 5 إلى 9 log cfu/g، كما أن الفاكهه المضافة لم تؤثر على حيوية بكتيريا البروبيوتك.

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