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Lactulose Production from Milk Permeate and its Performance in Healthy Functional Frozen Yoghurt

[1]R.A.Awad, [1]A.E. Hagrass, [2]Wafaa M. Salama,
[3]F.A. Abd Elmalek and [3]Amal I. Eldardiry

[1]Food Science Department, Faculty of Agriculture,
Ain Shams University, Cairo, Egypt
[2]Dairy Research Department, Food Technology Research Institute,
Agricultural Research Center, Giza, Egypt
[3]Dairy Chemistry Department, Animal Production Institute,
Agricultural Research Center, Dokki, Giza, Egypt

Abstract: Milk permeate was treated to increase its lactulose content by isomerization process of lactose. The produced high lactulose permeate (HLP) was used to manufacture frozen yoghurt using different starter cultures containing probiotic bacteria. Two control treatments of frozen yoghurt were made, one of them using regular permeate (RP) and the second using HLP. Each of them was inoculated with 3% yoghurt culture (YC). Three functional frozen yoghurt were made using HLP and inoculated with 1.5% YC+1.5% of *Lac. acidophilus* (T_1), *Lac. casei* (T_2) or *Bifido. bifidum* (T_3). The results showed a slight decrease in pH and higher total bacterial count during incubation period of all yoghurt treatments including controls. At the end of incubation, the probiotic count was higher in treatments with HLP. Lactose content was higher in control of RP than treatments of HLP. Functional treatments of HLP had lower freezing points and higher specific gravity, viscosity, acetaldehyde and diacetyle compared to control of RP. Incorporating HLP in the base formula of frozen yoghurt led to increase sp.gr and decrease overrun in resultant product being the lowest in treatment with *Lac. acidophilus* (T_1). Functional frozen yoghurt with HLP showed higher melting resistance than control of RP. Total and probiotic bacterial counts were higher in treatments with HLP compared to control of RP. Freezing process of yoghurt mix caused a slight and insignificant reduction in viable bacteria (total, probiotic) in all samples including controls, while frozen storage caused a significant reduction in survival of probiotic bacterial count. Functional frozen yoghurt with HLP was more preferable to panelists, while that with starter culture containing *Lac. acidophilus* (T_1) or *Bifido. bifidum* (T_3) were more preferable.

Key words: Frozen yoghurt · Lactulose · Probiotic · Synbiotic

INTRODUCTION

Functional foods have recently emerged as a novel sector of health-enhancing products and now recognized as foods for specified health use [1]. Such food provides a health benefit that goes beyond a general nutritional benefit. It can include those foods these are whole, fortified, enriched or enhanced, while neutralceuticals are considered as isolated components that can be incorporated into foods to enhance health at levels not usually obtainable from normal foods [2]. Lactulose (4-0-ß-D-galactopyranosyl-D-fructose) is an isomer of lactose in which the glucose aldehyde is converted to a ketone (fructose) by alkali hydroxide catalysis [3]. Lactulose is not found in milk, but is found in various whey and heated milk [4]. Lactulose syrup is a widely used pharmaceutically in Japan, US, and worldwide. It has a mildly purgative action and inhibits the growth of ammonia-producing organisms, thereby aiding in the treatment of chronic hepatoportal encephalopathy, a

Corresponding Author: Wafaa M. Salama, Dairy Research Department, Food Technology Research Institute, Agricultural Research Center, Giza, Egypt.

condition in which the brain is affected by nitrogenous substances from the colon [4, 5]. Lactulose is not digested in the small intestine but in large intestine, it promotes the growth of bifidobacterium species. This trait is especially of interest in nutrition of infants for whom lactulose is thought to stimulate growth of intestinal flora similar to those in breast-fed babies which bifidobacterium species are the predominant microorganisms [3, 6]. The role such bacteria is well known and also in making the probiotic food. Probiotic foods are defined as foods containing live microorganisms believed to enhance health by improving the balance of microflora in the gut [7]. Prebiotics are non-digestible dietary components that pass through the colon and selectively stimulate the proliferation and/or activity of probiotic bacteria in the colon [8]. Synbiotic is a product in which both probiotic and prebiotic are combined in a single product that beneficially affects the host by improving the survival and implantation of probiotic in gastrointestinal tract [9, 10]. Among dairy products with live cultures, probiotic ice cream and fermented frozen desserts, which gaining popularity [11].

During cheese production, the disposal of whey and milk permeate can be a source for environmental pollution. It could be estimated that more than 1.5 million tons of whey and milk permeate are generated in Egypt per year. More than 99% of this potential resource is discarded. Therefore, the present study was planned to provide a new technique for benefiting from milk permeate as a waste and environmental pollution by converting its lactose to lactulose which can be considered as prebiotic component. The ample formulations and properties of synbiotic dairy product using the prepared permeate syrup of high lactulose content with adding probiotic bacteria to produce acceptable functional frozen yoghurt were also described and investigated.

MATERIALS AND METHODS

Materials: Fresh permeate was obtained from dairy processing unit, Animal Production Research Institute, Agricultural Research Center, Giza, Egypt. Cream (60.2% fat, 3.7% SNF) was obtained by separation of buffalo's milk using Alfa Laval separator. Skim milk powder (medium heat) extra grade, (96.0% total solids) made in Holland was obtained from the local market. Commercial grade crystalline sucrose was obtained from Sugar and Integrated Industries Company, Giza, Egypt. Commercial Stabilizer/Emulsifier, EXN9080 was obtained from MIFAD (Misr Food Additives company), Egypt. Vanilla was obtained from the local market. Yoghurt starter culture (YC) consists of *Str. thermophilus* and *Lacto. bulgaricus* was obtained from Cairo MIRCEN Culture Collection Center, Faculty of Agriculture, Ain Shams University, Cairo, Egypt. Cultures of *Lac. acidophilus, Bifido. bifidum* and *Lac. casei* (DVS) were obtained from Chr. Hansen Lab., Copenhagen, Denmark. Activated carbon powder (Darco D60, 100 meshes) was obtained from El-Wattania for maize products company, 10^{th} of Ramadan city, Egypt. Anaerogen caches for anaerobic conditions and all microbiological media were obtained from Oxoid Ltd., Hampshire, England. Egg shell powder was prepared from white egg shells after washing with tap water to remove all adhering albumen, dried at 105°C/8h, ground in a ball mill mixer, (Mixer Mill MM200, Germany) at 800 rpm for 30 min and used as a catalyst.

High Lactulose Permeate (HLP) Preparation: Milk permeates of high lactulose content was prepared as described by Montilla *et al.* [12] with some modification as follows: Permeate was placed in round - bottom flask provided with an additional neck sampling inlet. Sodium hydroxide (4N) was added to permeate with agitation until the pH value reached 11. At this point, 40g/L of egg shell powder was added as a catalyst. The flask containing permeates and egg shell powder was immersed in a water bath at 70°C for 3h with periodically agitation. After first hour, 3% of activated carbon powder (charcoal) was added and left into mixture till the end of heating (isomerization) process after 2 additional hrs to discoloration. Egg shell was removed from solution by centrifugation at 20°C, 5000g for 10 min. Supernatant was filtrated using Whatman 1, collected and stored at 5°C until used.

Preparation of Functional Frozen Yoghurt: Frozen yoghurt mixes standardized to contain 8% fat, 15% sugar, 12% MSNF, 0.35%stabilizer/ emulsifier (S/E) and 0.01% vanilla were prepared and manufactured according to the procedure of Goda *et al*. [13] with some modification as follows: Freshly regular permeate (RP) or prepared high lactulose permeate (HLP) was firstly used to dissolve the required amount of skim milk powder and cream. The quantity was divided into two equal portions. The calculated required amounts of sugar and S/E were added to the first portion with continuous agitation during heat treatment at 85°C for 1 min, homogenized in duple stage homogenizer (RANNIE model 50.120H, Germany, 2500/500, lb) at same temperature, hold for 5 min, then cooled to 5°C and kept at the same temperature over night for aging. The second portion prepared of HLP was

Table 1: Characteristics of regular and high lactulose permeate used in preparation of functional yoghurt treatments

Character assessed	Regular permeate (RP)	High lactulose permeate (HLP)
pH value	6.50	7.00
Acidity (%)	0.22	0.18
Total solids (%)	5.40	6.31
NPN (%)	0.12	0.22
Ash (%)	0.51	0.94
Lactose (%)	4.62	0.31
Lactose reduction (%)	0.0	93.48

NPN = non protein nitrogen

Table 2: Composition (%) of functional frozen yoghurt recipes containing high lactulose and different starter cultures

	Treatments*				
	Controls				
Ingredients (Kg/100Kg mix)	RP	HLP	T1 (LA)	T2 (LC)	T3(BB)
Sugar	15	15	15	15	15
Stabilizer/ emulsifier	0.35	0.35	0.35	0.35	0.35
Cream	13.33	13.33	13.33	13.33	13.33
Skim milk powder	11.98	11.98	11.98	11.98	11.98
Starter culture	3 (YC)	3 (YC)	1.5YC +1.5LA	1.5YC +1.5LA	1.5YC +1.5BB
Regular permeate (RP)	56.34	-	-	-	-
High lactulose permeate (HLP)	-	56.34	56.34	56.34	56.34

*RP: regular permeate, HLP: high lactulose permeate, T1, T2, T3: treatments of HLP with *Lac. acidophilus*, *Lac. casei* and *Bifido. bifidum* as probiotic strains, respectively

heated at 85°C for 15 min, cooled to 45°C, and then divided into 4 parts before inoculation with different starter cultures to manufacture yoghurt as follows:

- T1 (control): inoculated with 3% yoghurt culture (YC) and served as control.
- T2 (LA): inoculated with 1.5% YC + 1.5 % *Lac. acidophilus*.
- T3 (LC): inoculated with 1.5% YC + 1.5 %. *Lac. casei*.
- T4 (BB): inoculated with 1.5% YC + 1.5 *Bifido. bifidum*.

Another control treatment was manufactured of RP base mix and inoculated with 3% YC. After inoculation, all treatments were incubated at 42°C until reaching pH 4.6, then transferred to refrigerator at 5°C .All fermented parts were analyzed during fermentation period for acid production, pH value, total bacterial count and specific microorganisms. Frozen yoghurt treatments were done by mixing one part of first portion (base mix) with equal part of second fermented portion (to mend product) Vanilla was added to each mix before freezing in a horizontal batch freezer (Qutofrigor, E.21.8, Co., France). The frozen yoghurt treatments were packaged in plastic cups (80 ml) and hardened at -26±2°C /24 hrs before storage at -18°C. Samples were taken of fresh yoghurt mixes after well combining and frozen products to assess the product properties either when fresh or during storage. Compositional analysis of RP and HLP are shown in Table 1 while the mix composition of frozen yoghurt recipes with different starter cultures is shown in Table 2.

Methods of Analyses: Moisture, ash, and protein contents were determined according to A.O.A.C [14]. Fat content was determined according to Ling [15]. Titratable acidity was determined according to Richardson [16]. Values of pH were measured using pH meter (Hanna model 8417). Lactose content was determined by the method of Lawrance [17]. Acetaldehyde and diacetyl contents were estimated according to Lee and Jago [18]. Total viable bacterial counts were enumerated according to the method described by Houghtby *et al.* [19]. Bacterial strains of *Bifido. bifidum* and *Lac. acidophilus* were enumerated according to Dave and Shah [20] while *Lac. casei* was counted as reported by Vinderola and Reinheiner [21]. Yoghurt organisms (*Str. thermophilus, Lac. bulgaricus*) were determined as described by Lee *et al*. [22]. Specific gravity values of mixes and resultant ice cream were determined at 20°C using the pycnometer according to Winton [23]. The overrun percentages in resultant frozen yoghurt were assessed according to Marchal *et al*. [24]. The freezing point was tested for the mixes as in FAO [25]. Melting resistance of frozen yoghurt was examined according to Reid and Painter [26]. Dynamic viscosity of base mixes was determined as described by Toledo [27] using Brookfield viscometer (Brookfield RV-HA-HB model, Brookfield Engineering Laboratories, Inc., USA) equipped with a spindle No SC4-21. Samples of functional frozen yoghurt were organoleptically judged by a panel of 10 judges from Food Sci. Dept., Fac. Agric., Ain Shams Univ. and Dairy Division, Animal Production Res. Inst. according to Arbuckle [28]. All achieved data were statistically analyzed using the General Linear Models procedure of the Statistical Analysis System SAS [29] and significance was defined at $p \leq 0.05$. All experiments as well as the related analyses were repeated three times and all obtained data are expressed as averages.

RESULTS AND DISCUSSION

Properties of Bio-Yoghurt with High Lactulose Content:

Values of pH and Titratable Acidity: Changes in pH and titratable acidity of functional bioyoghurt with high lactulose content and probiotic bacteria are shown in Table 3. The data indicated that the pH value decreased and acidity increased in all treatments including controls during incubation period. At the end of incubation, control of RP had the highest pH with lowest acidity. This means that the growth of starter microorganisms was slower in control of RP and may be due to the presence of lactulose which may act as a prebiotic component and stimulate the growth of starter microorganisms. At the end of incubation period, functional yoghurt treatment with starter culture containing *Lac. acidophilus* (T_1), exhibited the lowest pH (4.7) with highest acidity (0.74%) among all treatments. This means that the activity and growth of starter culture was highly stimulated with lactulose.

Total and Probiotic Bacterial Counts: Total and probiotic bacterial counts (log cfu/ ml) of all samples are stated in Table 4. Total bacterial count increased during incubation by about 2 log cfu/ ml in all treatments. At the end of incubation, the probiotic count was higher in treatments with HLP including control by almost 2 log cycle. This could be attributed to the presence of lactulose as an activator.

Properties of Functional Frozen Yoghurt Mixes: Properties of the prepared functional frozen yoghurt mixes are presented in Table 5. Total solid, protein and fat contents were in narrow range since there were previously adjusted in the recipes. Lactose percentage was higher in control treatment of RP (4.91%) while all treatments with HLP had lower values. On the other hand, ash content was lower in control treatment of RP (1.42%) while it was higher in treatments with HLP. The lower lactose and higher ash contents in treatments with HLP are mainly due to their original contents and process of the prepared permeate with high lactulose (Table 1). The acidity increased with the presence of lactulose and probiotic

Table 3: Changes in pH value and titratable acidity of functional yoghurt with high lactulose content and different probiotic cultures during incubation period.

	Treatments*				
	Controls				
Incubation time (min)	RP	HLP	T1 (LA)	T2 (LC)	T3 (BB)
pH value					
0	6.5	6.4	6.5	6.5	6.5
30	6.2	6.1	6.3	6.2	6.2
60	6.0	5.9	6.1	5.9	5.8
90	5.7	5.7	5.8	5.7	5.5
120	5.4	5.4	5.5	5.5	5.3
150	5.3	5.0	4.9	5.0	5.1
180	5.1	4.8	4.7	4.8	4.9
Acidity (%)					
0	0.20	0.21	0.22	0.22	0.22
30	0.23	0.24	0.25	0.25	0.24
60	0.31	0.33	0.35	0.35	0.32
90	0.40	0.42	0.45	0.44	0.42
120	0.49	0.52	0.55	0.53	0.49
150	0.5 7	0.61	0.64	0.62	0.60
180	0.64	0.69	0.74	0.71	0.66

*RP: regular permeate, HLP: high lactulose permeate, T1, T2, T3: treatments of HLP with *Lac. acidophilus*, *Lac. casei* and *Bifido. bifidum* as probiotic strains, respectively

Table 4: Total and probiotic bacterial counts (log cfu/ml) of functional yoghurt with high lactulose content and different probiotic cultures during incubation period

		Treatments*				
		Controls				
Type of strain	Incubation time (min)	RP	HLP	T1 (LA)	T2 (LC)	T3 (BB)
Total bacterial count	After inoculation (0.0)	4.49^{a}	4.48^{a}	4.19^{b}	4.21^{b}	4.19^{b}
	End of incubation (180)	6.36^{a}	6.39^{a}	6.24^{b}	6.25^{b}	6.23^{b}
Probiotic bacterial count	After inoculation (0.0)	5.32^{a}	5.38^{a}	4.27^{b}	4.26^{b}	4.25^{b}
	End of incubation (180)	6.95^{c}	8.02^{b}	8.61^{a}	8.61^{a}	8.60^{a}

* See legend to Table 3 for details.

abc: Means with the same letter among treatments are not significantly different.

Table 5: Mix Properties of functional frozen yoghurt with high lactulose content and different probiotic starter cultures.

	Treatments*				
	Controls				
Properties	RP	HLP	T1 (LA)	T2 (LC)	T3 (BB)
Total solids %	36.82	36.71	36.73	36.70	36.72
Fat %	8.20	8.21	8.20	8.23	8.20
Protein %	5.71^{b}	5.83^{a}	5.85^{a}	5.81^{a}	5.85^{a}
Lactose %	4.91^{a}	3.17^{c}	3.27^{b}	3.17^{c}	3.17^{c}
Ash %	1.42^{b}	1.79^{a}	1.78^{a}	1.78^{a}	1.79^{a}
Acidity%	0.40^{c}	0.42^{ab}	0.46^{a}	0.43^{ab}	0.41^{bc}
pH value	5.71^{a}	5.67^{a}	5.51^{b}	5.66^{a}	5.67^{a}
Acetaldehyde (μg/100ml)	126^{d}	134^{c}	145^{ab}	142^{bc}	148^{a}
Diacetyl (μg/100ml)	70^{b}	77^{a}	79^{a}	76^{a}	78^{a}
Specific gravity	1.1202^{e}	1.1302^{d}	1.1353^{a}	1.1312^{c}	1.1330^{b}
Freezing point (°C)	-2.51^{c}	-2.68^{b}	-2.83^{a}	-2.80^{a}	-2.74^{ab}

* See legend to Table (3) for details abc: Means with the same letter among treatments are not significantly different

bacteria in the base mix. The presence of lactulose may act as a prebiotic material that enhance the starter activity and developed acidity. It was highest in treatment with starter culture containing *Lac. acidophilus* and was the lowest in control treatment of RP. The pH values followed an opposite trend to that of acidity.

The acetaldehyde and diacetyl were higher in treatments with HLP due to the higher activity and survival number of probiotic bacteria in those treatments. Specific gravity (Sp.gr) was lower in control of RP than that of HLP. This may be due to the Sp.gr of raw permeate since the HLP contains higher TS. Frozen yoghurt mixes made of HLP had lower freezing points than that of RP. This is mainly due to the more hydrolysis and fermentation of lactose in HLP to other several components that have lower molecular weight. The viscosity of all mixes (Fig. 1) increased by increasing the shear rate value, which means that the yoghurt mixes behave as a pseudoplastic material. Control treatment of RP showed lower viscosity than that of treatments with HLP. The higher viscosity value of HLP treatments could be due to the higher viscosity of permeate used and the higher acidity in the mix as a result of higher starter activity. T_3 (LA) of HLP showed the highest viscosity value among all treatments since it had the highest acidity.

Properties of Resultant Frozen Yoghurt: Specific gravity (Sp.gr) of functional frozen yoghurt was affected by using RP or HLP in the formula. The data in Table 6 indicated that control treatment of RP had lower sp.gr than that of all treatments with HLP while there were slight differences in sp.gr values among treatments of high lactulose contents. Several factors would affect sp.gr such as the mix composition, viscosity, and state of protein. Incorporating high lactulose permeate in the base formula led to increase sp.gr. This could be due to the higher TS, viscosity and lower freezing point of base mix. The overrun percentage was higher in control treatment of RP than that in all treatments of HLP with any starter culture. This could be due to the corresponding lower sp.gr and higher freezing point. The overrun percentages of resultant symbiotic frozen yoghurt are affected by different factors such as the state and nature of proteins,

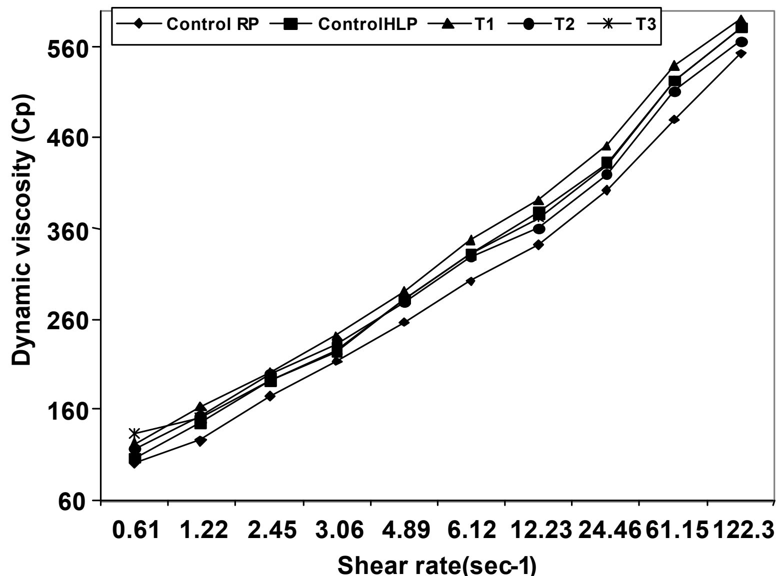

Fig. 1: Dynamic viscosity of functional frozen yoghurt mixes with high lactulose contents and different probiotic starter cultures.

Table 6: Some properties of functional frozen yoghurt with high lactulose content and different probiotic starter cultures

	Treatments*				
	Controls				
Character assessed	RP	HLP	T1 (LA)	T2 (LC)	T3 (BB)
Specific gravity	0.6462^{c}	0.6650^{b}	0.6790^{a}	0.6700^{ab}	0.6740^{a}
Overrun %	73.35^{a}	69.95^{ab}	67.20^{b}	68.84^{b}	68.10^{b}
Loss percentage after (min)					
15	11.5	9.5	8.2	9.0	9.5
30	37.3	24.5	22.7	23.4	24.0
45	79.2	58.6	48.6	50.9	51.7
60	89.3	81.1	78.8	80.3	79.8
75	96.4	92.2	87.2	90.7	90.1
90	98.7	97.9	97.3	98.0	98.1

*See legend to Table 3 for details abc: Means with the same letter among treatments are not significantly different

acidity and freezing point of base mixes. The obtained data are in agreement with Salem *et al*. [30] who found that adding fermented milk with probiotic bacteria caused a decrease in overrun percentage. Among treatments with HLP, there was a slight difference in overrun being the lowest in treatment with *Lac. acidophilus* as a part of starter culture (T_1). The lowest overrun in T_1 could be due to the higher acidity and lower freezing point. The melting behavior of samples with high lactulose contents and probiotic starter culture (Table 6) indicated that all samples with HLP were slower in melting and showed higher melting resistance than that of control with RP. The data also stated that all treatments with HLP had almost similar melting resistance. The melting behavior of frozen yoghurt and ice cream products is mainly affected by molecular weight and type of soluble component in the base mix which by its turn affect the freezing point.

Micorobiological Examination: Control of RP had lower total bacterial count than that of treatments with high lactulose contents (Table 7). This is mainly due to the presence of lactulose which is known as prebiotic material and therefore encourage and improve the growth and survival of bacteria. The freezing process of yoghurt mix caused a slight reduction in viable bacteria of all samples including controls. All samples during frozen storage, exhibited lower bacterial count than that of initial count after freezing being lowest in control of RP. After 3

Table 7: Total viable bacterial count and survival of probiotic strains (log cfu/ml) in synbiotic frozen yoghurt during processing and frozen storage at -20 °C up to 3 months.

	Treatments*				
	Controls				
Storage period (month)	RP	HLP	T1 (LA)	T2 (LC)	T3 (BB)
Total bacterial count					
Fresh mix (before freezing)	6.39	7.42	7.38	7.39	7.38
After freezing (0.0)	6.35	7.38	7.33	7.35	7.34
1	6.11	6.65	6.84	6.73	6.93
2	5.79	6.48	6.50	6.49	6.59
3	5.48	6.21	6.24	6.22	6.23
Probiotic bacterial count					
Fresh mix (before freezing)	7.22	8.35	9.64	9.62	9.63
After freezing (0.0)	7.19	8.33	9.62	9.35	9.62
1	7.00	8.21	9.60	9.35	9.60
2	6.72	8.07	9.24	9.13	9.33
3	6.02	7.70	8.62	8.49	8.81

*See legend to Table (3) for details

Table 8: Scoring of functional frozen yoghurt attributes made with high lactulose content and different probiotic starter.

		Sensory attributes				
Treatments*	Storage period (month)	Flavor (45)	Body& texture (35)	Melting properties (10)	Appearance (10)	Total Score (100)
Control (RP)	Fresh	39	29	9	8	85[hg]
	1	38	30	8	8	84[hi]
	2	39	28	8	8	83[i]
	3	37	27	7	7	78[j]
Control (HLP)	Fresh	41	32	9	9	91[c]
	1	40	30	8	9	87[ef]
	2	39	30	9	8	86[fg]
	3	38	29	8	8	83[i]
T_1 (LA)	Fresh	43	33	9	9	94[a]
	1	43	32	9	9	93[ab]
	2	42	33	9	9	92[bc]
	3	40	30	8	8	86[fg]
T_2 (LC)	Fresh	41	32	9	9	91[c]
	1	41	31	9	8	89[d]
	2	40	30	9	9	88[de]
	3	38	29	8	8	83[i]
T_3 (BB)	Fresh	43	31	10	9	93[ab]
	1	42	32	9	9	92[bc]
	2	41	32	9	9	91[c]
	3	38	29	8	8	83[i]

*See legend to Table (3) for details abc: Means with the same letter among treatments are not significantly different

months of storage at -20°C frozen yoghurt treatments of high lactulose content (HLP) showed almost close viable bacterial count but still higher than control of RP. The data in Table 7 also indicated that there was a reduction by about one log cycle in viable bacterial count of all synbiotic frozen yoghurt samples after 3 months of frozen storage. These findings are in agreement with Salem *et al*. [30].

Survival of probiotic bacteria (Table 7) stated that probiotic bacterial count in control mix treatment of RP was lower than that in other mixes with HLP. The count number showed slight and non- significant decrease after freezing process in all yoghurt treatments including controls. Frozen storage of functional synbiotic yoghurt caused a significant reduction in survival of probiotic bacterial count. The effect was more noticeable after 3 months of storage at -18°C. Such reduction was noticed in all treatments including controls by about one log cycle. Meanwhile, the number of probiotic bacteria was still higher than 8 log cfu/ml which is the lowest limit of number that recommended in several researches to be as a probiotic product. The results obtained are in agreement with Bednarski *et al*. [31] and Tabatabaie and Mortazavi [32], who found a stimulating effect and survival improvement of lactulose on *Bifido. bifidum*. Incorporating probiotic bacteria into the culture of frozen yoghurt containing high lactulose contents increased the probiotic counts. Frozen storage had significant effect on reduction of probiotic counts and *Lac. casei* was the mostly affected culture.

Organoleptic Properties: Sensory evaluation of the prepared product is shown in Table 8. Treatments with high lactulose contents possessed better flavor than control RP. Among all treatments, the one contain either *Lac. acidophilus* or *Bifido. bifidum* gained the best flavour. There were some improvements in body& texture of frozen yoghurt samples manufactured using HLP since all scored higher points than that of control with regular permeate (RP). Frozen yoghurt samples were smoother with less iciness by increasing lactulose content in the base mix. Melting properties and appearance of functional frozen yoghurt with high lactulose content were much better than that of RP. Treatment contains Bifido. *bifidum* (T_3) was the best in melting quality and appearance among all synbiotic frozen yoghurt samples. The data also indicated that increasing lactulose content in base formula produced more preferable samples to panelists than that of regular permeate. The samples produced with starter culture containing *Lac. acidophilus* (T1) or *Bifido. bifidum* (T3) were the most preferable. All sensory attributes scored lower points during storage at -18°C. This agrees with Salem *et al*. [30]. In conclusion, functional synbiotic frozen yoghurt with high lactulose content can be produced using milk permeate after conversion process of lactose to lactulose. The yoghurt can be inoculated with a mixture of yoghurt culture and probiotic bacteria. All functional frozen yoghurt samples were acceptable but the samples with starter culture containing *Lac. acidophilus* or *Bifido. bifidum* were more preferable to panelists.

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