

## Manufacture and Properties of Low-Fat Bio Yoghurt Containing Probiotic Strains and Maltodextrin as Prebiotic

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### Abstract

**Background and Objective:** Significant research has been focused in discovering which prebiotics is most beneficial in increasing the levels of probiotic bacteria in the gastrointestinal tract. Therefore, the present study was carried out to investigate the effect of adding maltodextrin as prebiotics on the properties and survival of different probiotic strains in low-fat bio-yoghurt during cold storage.

**Methodology:** Low-fat bio-yoghurt was made using probiotic strains (*Lb. acidophilus* NCTC12980R and *Bifidobacterium bifidum* NCTC1300R) and 2% maltodextrin as well as compared with traditional starter (*Str. thermophilus* and *Lb. delbrueckii* ssp. *bulgaricus*). Bio-yoghurt samples were evaluated for chemical, microbiological and organoleptic properties during storage at 4°C for 21 days.

**Results:** The type of starter culture used did not affect on the dry matter, ash contents and viscosity in different bio-yoghurt. Culture combinations, fortification of maltodextrin and storage period significantly influenced on the acidity, SN\TN, diacetyly, acetaldehyde contents and viscosity in different bio-yoghurt. The results showed that maltodextrin had no significant effect on the viability of yoghurt cultures and *Lb. acidophilus* strain while, it stimulated the growth of *Bifi. bifidum* starter bacteria to a great extent. Generally, the counts of probiotic strains used in bio-yoghurt made with maltodextrin were still higher than the recommended minimum levels ( $10^7$  cfu/ ml) along the storage period.

**Conclusion:** Addition of maltodextrin and use of probiotic strains such as *Lb. acidophilus* and *Bifi. bifidum* enhanced the sensory properties of bio-yoghurt samples when fresh and along the storage period.

**Keywords:** Bio-yoghurt; Probiotics; Maltodextrin; Prebiotic

### Introduction

Functional foods have recently emerged as a novel sector of health-enhancing products. The target of functional foods is largely dependent on the ingredients used. The concept of functional foods has evolved as the role of food in the maintenance of health. Yoghurt is one of the best-known of the dairy products that contain viable lactic acid bacteria. Yoghurt is defined by the Codex Alimentarius of 2003 as a coagulated milk product that results from the fermentation of lactic acid in milk by *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *Bulgaricus* [1]. Therefore, this type of bacteria called yoghurt starter culture. These bacteria are not bile acid resistant and do not survive in the passage of intestinal tract. Thus, recently, probiotic bacterial strains such as *Lb. acidophilus* and *Bifidobacterium* ssp., incorporated into yoghurt starter culture due to their bile-resistant properties and beneficial health effects. The resulting product, called as “yoghurt-like products”, “probiotic” or “bio-yoghurts”, are becoming more popular due to the ability of excellent health effects of probiotic bacteria [2]. Probiotic bacteria are defined as living microorganisms administered in a sufficient number to survive in the intestinal ecosystem, and must have a positive effect on the host [3]. As the viability of live probiotic bacteria in food products and among transit

through the gastrointestinal tract may be variable, the prebiotic concept has been developed. The efficiency of added probiotic bacteria depends on dose level and their viability must be maintained throughout storage, and they must survive the gut environment [4-6]. In order to improve these features of probiotic bacteria, fermented food should be supplemented with prebiotics. There are non-digestible food ingredients that beneficially affects the host by selectively stimulating the growth and/or activity of one or a limited number of bacteria in the colon that can improve host health [7]. Thus, the prebiotic approach advocates administration of non-viable entities and aims to enhance survival of probiotics in the upper gastrointestinal tract. Certain polysaccharides which cannot be digested, except through probiotic bacterial activity, are prebiotics. Those that contain fructose (e.g., inulin) are able to alter the composition of human gut flora towards a predominance of probiotic bacteria. There has been a considerable interest in the use of some polysaccharides as prebiotics to enhance the survivability and colonization of probiotic bacteria added in food products. Because of the difficulty in maintaining a probiotic in the gastrointestinal tract, significant research has been focused in discovering which prebiotics is most beneficial in increasing the levels of probiotic bacteria in the gastrointestinal tract [8]. Recently, maltodextrin have been included in the prebiotic group owing to their indigestibility properties. Maltodextrins are maltooligosaccharides with a degree of polymerization ranging from three to nine and often

act as flavour enhancers, fat replacers and bulking agents in dairy product [9]. Yoghurt products with a low fat content aimed for reducing the daily energy intake and hence for improving the energy balance may lack the mouth-feel and texture associated with higher fat products. Several researchers have described the effects of fat replacer such as inulin and maltodextrin on the sensory quality of low fat fermented milk [10]. Therefore, additionally the prebiotic properties of maltodextrin to stimulate the growth and activity of probiotic strain, it could be used to improve the texture and taste of low-fat yoghurt.

Therefore, the aim of the present study was to attempt the production and characterization a new type of low-fat bio-yoghurt made with yoghurt culture and different probiotic strains fortified with maltodextrin for enhancing nutritional and functional values of this product.

## Materials and Methods

### Ingredients

Fresh buffalo's milk was obtained from the herd of the dairy cattle at Faculty of Agriculture, Ain Shams University, Egypt. Skim milk powder (97% DM) made in Poland was obtained from the local market of Cairo. Maltodextrine (Spray-dried product obtained by enzymatic conversion of corn starch) was obtained from National Company for Maize Products, 10 of Ramadan City, Industrial Zone A1, El Sharkia, Egypt.

### Starter cultures

Yoghurt cultures used in this study were commercially named YC-X11 DIP 50 u consists of (*Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *Bulgaricus* 1:1) and generally used for yoghurt fermentation. Commercial freeze-dried bacterial starter was obtained from Chr. Hansens Laboratiers, Denmark and prepared as the mother culture by adding 1% of lyophilized cell culture into 12% sterilized reconstituted skim milk and incubated at 42°C for 4-6 h before 24 h from using. Two probiotic strains provided by Quality Medical Sciences Co. Ltd, were used in this study includes *Lb acidophilus* NCTC12980R and *Bifi. bifidium* NCTC1300R. Each strain was propagated in MRS broth medium supplemented with 0.05% cystein hydrochloride at 37°C for 24 h. Stock cultures of probiotic strains were made by mixing a pure culture that had been grown over night with equal amount of solution and stored at -20°C until experimentally used. Mother culture was prepared by adding 1% of stock culture into 12% sterilized reconstituted skim milk and incubated at 37°C for 7-8 hrs before 24 h from using.

### Production of different bio-yoghurt samples

Fresh buffalo's milk was skimmed and standardized to 0.5% fat and 14% TS using skim milk powder. The standardized milk was divided into five portions. Two portions (T2 and T4) were enriched with 2% maltodextrin as a prebiotic. All the milks were heated at 90°C for 10 min subsequently cooled to 42°C. To manufacture of different bio-yoghurt, the five portions of heat treated milk inoculated with different starter cultures as follows:

Control: Liquid skim milk without maltodextrin inoculated with 3% yoghurt starter culture

T1: Liquid skim milk without maltodextrin inoculated with 1.5% yoghurt culture+1.5% *Lb. acidophilus*

T2: Liquid skim milk with 2% maltodextrin inoculated with 1.5% yoghurt culture+1.5% *Lb acidophilus*

T3: Liquid skim milk without maltodextrin inoculated with 1.5% yoghurt culture+1% *Bifi. bifidium*

T4: Liquid skim milk with 2% maltodextrin inoculated with 1.5% yoghurt culture+1.5% *Bifi. Bifidium*

All treatments were incubated at 42°C till coagulation (pH 4.7) then cooled to 4°C. Three replicates were done for every treatment. The resultant fermented milks were stored at 4°C for 21 days. Samples were taken when fresh and after 3, 7, 14 and 21 days of cold storage and analyzed chemically, microbiologically, and organoleptically.

### Chemical composition of bio-yoghurt

Dry matter, ash, titratable acidity as lactic acid (TA), total and soluble nitrogen contents were determined in yoghurt samples by the method described in AOAC [11]. Acetaldehyde and diacetyl contents were determined in yoghurt samples according to Lees and Jago [12,13] using Conway micro diffusion-Semi carbazide method.

### Apparent viscosity

Viscosity of different yoghurt samples was measured using a rotational coaxial viscometer (RHEOTEST II-Medingen, Germany) at shear rates ranging from 1.000 to 437.4 sec<sup>-1</sup> according to Toledo [14]. The measuring device (S2) was used and samples adjusted to 20 ± 1 before loading in the viscometer device. Apparent viscosity (cp) of different samples was calculated at share rate 145.8 s<sup>-1</sup>.

### Microbiological examination of bio-yoghurt products

*Bifidobacteria* were enumerated according to Dave and Shah [15] using modified MRS agar supplemented with 0.05% L-cystein and 0.3% lithium chloride. The plates were anaerobically incubated at 37°C for 48-72 h. *Lb. bulgaricus* count was determined using MRS agar (pH, 5.2) according to Tharmaraj and Shah [16]. The plates were anaerobically incubated at 43°C for 72 h. On the other hand *Lb. acidophilus* count was determined using Bile MRS Agar according to Vinderola and Reinheimer [17]. The plates were aerobically incubated at 73°C for 72 h. *Str. thermophilus* count was determined using M17 agar medium [18]. The plates were aerobically incubated at 37°C for 48 h. Coliform count was enumerated using Violet Red Bile Agar medium as reported by American Public Health Association [19]. The plates were incubated at 37°C for 48 h. Yeasts and moulds were determined on Malt-Extract Agar medium as suggested by Harrigan and McCance [20]. The plates were incubated at 25-27°C for 4 days.

### Organoleptic evaluation of bio-yoghurt

The organoleptic properties of different bio-yoghurt samples were assessed by regular taste panel of the staff-member at Food Science Department, Faculty of Agriculture, Ain Shams University. Yoghurt samples were evaluated for flavor (60 points), body and texture (30 points) and appearance (10 points) according to Bodyfelt et al. [21].

### Statistical analysis

Statistical analysis was performed according to SAS Institute [22] using General Linear Model (GLM) with main effect of treatments. Duncan's multiple range was used to separate among means of three replicates at P ≤ 0.05.

## Results and Discussion

### Chemical composition of bio-yoghurt

As shown in Table 1, the type of starter culture used did not affect on the dry matter content in different bio-yoghurt. While adding maltodextrin as prebiotic to different bio-yoghurt caused a clear significant increase in dry matter. Dry matter content slightly increased in all bio-fermented milk treatments as the refrigerated storage period progressed up to 21 days. This increase in dry matter along the storage period may be due to water evaporation along the storage period [23,24]. It could be noticed that, the ash contents were not significant differences in the all treatments and control sample along the storage period. This means that the type of starter culture and addition of maltodextrin had no significant effect on the ash content in bio-yoghurt product. The slight differences could be observed in ash content of bio-yoghurt in all treatments during the storage period. These differences may be due to the changes in dry matter along the storage period. Generally, the ash content varied from 0.775% to 0.789% in fresh bio-yoghurt samples which has been slightly increased to 0.783 to 0.796 at the end of storage period. These differences may be associated with the dry matter changes along the storage period.

The results revealed that, control yoghurt sample had higher titratable acidity as compared with all other treatments. On the other hand, bio-yoghurt fermented with 1.5% yoghurt cultures+1.5% *Bifido. bifidum* without maltodextrin (T4) had lower titratable acidity compared with the other treatments. It could be noticed that, the level of acidity in different bio-yogurts was found to be lower than control yogurt. These results were in agreement with Ozer et al. Guler, Singh et al. and Ranathunga [25-28], who found that, traditional yoghurt fermented with yoghurt cultures was higher acidity and lower pH value than that bio-yoghurt made with different probiotic bacteria. Adding the maltodextrin to bio-yoghurt caused significant increase in the titratable acidity. These could be due to the effect of maltodextrin on the growth and/or activity of some lactic acid and probiotic bacterial starter cultures [29]. These results agree with Yeo and Liang [30] who stated that, supplementation the fermented soy milk with maltodextrin increased ( $P < 0.05$ ) the production of lactic acid. Also, Raju and Pal [31] observed that different bulking agents such as maltodextrin had a significant effect ( $P < 0.01$ ) on the acidity of artificial sweetened Mistidahi, and there were a significant ( $P < 0.01$ ) increase with maltodextrin compared to control. Generally, the titratable acidity gradually increased in all treatments along the storage period, this may be due to the activity of fermented milk cultures. The increase in the titratable acidity along the storage of yoghurt were also reported by Mehanna et al. Kebary et al. El Batawy and El Batawy et al. [24,32-34]. It appears that the composition of starter culture, addition of maltodextrin and storage period could be effected the overall level of acidity stored yoghurt samples [10].

It could be observed that, SN/TN content (%) gradually increased in samples as the storage period progressed. Moreover, there were nonsignificant differences in SN/TN content (%) among the control sample and other treatments containing *Lb. acidophilus* with or without maltodextrin (T1 and T3) when fresh and along the storage period. SN/TN content (%) was significantly lower in bio-yoghurt samples containing *Bifido. bifidum* with or without maltodextrin (T2 and T4) compared with other treatments. This is could be due to the

lower proteolytic activity of *Bifido. bifidum* compared with yoghurt and *Lb. acidophilus* cultures. The results are agreement with results obtained with Shihata; Donkor et al. [35,36] who stated that, proteolytic activity of *S. thermophilus*, *L. delbrueckii* ssp. *bulgaricus*, and *L. acidophilus* to be much greater than that of *Bifidobacterium* spp. Addition of 2% maltodextrin to bio-yoghurt caused non-significant increase in SN/TN content in final product.

Diacetyl and acetaldehyde are the main volatile compounds responsible for the aroma and play a considerable role in flavour development in fermented milk products during storage period. As shown in Table 1, nonsignificant differences were observed in diacetyl and acetaldehyde content between control yoghurt sample and other treatments containing *Lb. acidophilus* with or without maltodextrin (T1 and T3) when fresh and along the storage period. Moreover, diacetyl and acetaldehyde contents were significantly higher in bio-yoghurt samples containing *Bifi. bifidum* starter culture with or without maltodextrin (T2 and T4) compared with other treatments. The higher amount of diacetyl and acetaldehyde might be due to the metabolism of *bifidobacteria*. This means, culture combinations between yoghurt culture and *Lb. acidophilus* had no affected on diacetyl and acetaldehyde contents in bio-yoghurt. Therefore, using probiotic *bifidobacteria* in bio-yoghurt production caused a significant increase in diacetyl and acetaldehyde contents compared with control sample (yoghurt made with traditional yoghurt culture). These results are in agreement with those obtained by Yuguchi et al. [37] they found that, diacetyl and acetaldehyde contents were higher in fermented milk samples with combination of *Lb. delbrueckii* ssp. *bulgaricus*, *Str. thermophilus* and *Bif. longum* than the use of one strain alone. It was reported that *Lb. acidophilus* had lower diacetyl and acetaldehyde synthesising capacity than *bifidobacteria* [38]. Also, Barona et al. [39] found that, the fermented milk with no *bifidobacteria* contained less acetaldehyde than those that contained *bifidobacteria*. On the other hand, addition of 2% maltodextrin in bio-yoghurt containing yoghurt culture and *Lb. acidophilus* did not affect on the diacetyl and acetaldehyde contents in final bio-yoghurt. While adding maltodextrin as prebiotic to bio-yoghurt samples fermented with yoghurt culture and *Bifi. bifidum* caused a clear significant increase in diacetyl and acetaldehyde contents. This is may be due to the maltodextrin did not affect the growth and activities of yoghurt and *Lb. acidophilus* starter bacteria, but stimulated the growth of *Bifi. bifidum*. These results agree with Biser et al. [10] who stated that, addition of polysaccharides may enhance the starter activity specially *Bifido. bifidum*.

Acetaldehyde content gradually decreased in all low-fat yoghurt samples as the cold storage period progressed. While, diacetyl content increased till the 3rd day of the storage period followed by gradual decrease till the experimental end (21 days). The decrease in acetaldehyde content during the storage period is presumably due to the demonstrated ability of some starter culture to reduce acetaldehyde to ethanol or oxidize it to acetic acid as reported by Salama and Roushdy et al. [40,41]. The decrease in diacetyl mostly be due to slow reduction of diacetyl to acetone as detected by Diressen; Roushdy et al. [41,42].

Treatment	Storage period (day)				
	Fresh	3	7	14	21
<b>Dry matter%</b>					
C	13.94 <sup>Bd</sup>	14.06 <sup>Bd</sup>	14.15 <sup>Bc</sup>	14.38 <sup>Bb</sup>	14.46 <sup>Ba</sup>
T1	14.04 <sup>Bc</sup>	14.05 <sup>Bc</sup>	14.27 <sup>Bb</sup>	14.32 <sup>Bab</sup>	14.45 <sup>Ba</sup>
T2	15.95 <sup>Ac</sup>	16.03 <sup>Abc</sup>	16.23 <sup>Ab</sup>	16.39 <sup>Aa</sup>	16.42 <sup>Aa</sup>
T3	13.98 <sup>Bc</sup>	14.05 <sup>Bc</sup>	14.31 <sup>Bb</sup>	14.41 <sup>Ba</sup>	14.48 <sup>Ba</sup>
T4	16.02 <sup>Ad</sup>	16.10 <sup>Ad</sup>	16.27 <sup>Aac</sup>	16.36 <sup>Ab</sup>	16.50 <sup>Aa</sup>
<b>Ash%</b>					
C	0.775 <sup>Ab</sup>	0.776 <sup>Ab</sup>	0.783 <sup>Aa</sup>	0.784 <sup>Aa</sup>	0.789 <sup>Aa</sup>
T1	0.780 <sup>Ab</sup>	0.784 <sup>Ab</sup>	0.785 <sup>Aab</sup>	0.791 <sup>Aa</sup>	0.793 <sup>Aa</sup>
T2	0.778 <sup>Ab</sup>	0.780 <sup>Ab</sup>	0.782 <sup>Aab</sup>	0.788 <sup>Aa</sup>	0.792 <sup>Aa</sup>
T3	0.781 <sup>Ab</sup>	0.786 <sup>Ab</sup>	0.788 <sup>Aab</sup>	0.792 <sup>Aa</sup>	0.796 <sup>Aa</sup>
T4	0.783 <sup>Ab</sup>	0.784 <sup>Ab</sup>	0.789 <sup>Aab</sup>	0.792 <sup>Aa</sup>	0.794 <sup>Aa</sup>
<b>Titrateable acidity (%)</b>					
C	0.85 <sup>Ad</sup>	0.89 <sup>Ad</sup>	0.95 <sup>Ac</sup>	1.04 <sup>Ab</sup>	1.14 <sup>Aa</sup>
T1	0.74 <sup>Bd</sup>	0.76 <sup>Bd</sup>	0.82 <sup>Bc</sup>	0.88 <sup>BCb</sup>	0.95 <sup>BCa</sup>
T2	0.76 <sup>Bd</sup>	0.79 <sup>Bd</sup>	0.85 <sup>Bc</sup>	0.92 <sup>Bb</sup>	0.99 <sup>Ba</sup>
T3	0.70 <sup>Cd</sup>	0.73 <sup>Cd</sup>	0.78 <sup>Cc</sup>	0.82 <sup>Cb</sup>	0.91 <sup>Da</sup>
T4	0.76 <sup>Be</sup>	0.80 <sup>Bd</sup>	0.86 <sup>Bc</sup>	0.92 <sup>Bb</sup>	0.98 <sup>Ca</sup>
<b>SN/TN (%)</b>					
C	7.17 <sup>Ad</sup>	7.95 <sup>Ad</sup>	8.84 <sup>Ac</sup>	9.79 <sup>Ab</sup>	10.94 <sup>Aa</sup>
T1	7.01 <sup>Ad</sup>	7.66 <sup>Ad</sup>	8.54 <sup>Ac</sup>	9.49 <sup>Ab</sup>	10.54 <sup>Aa</sup>
T2	7.09 <sup>Ad</sup>	7.76 <sup>Ad</sup>	8.60 <sup>Ac</sup>	9.57 <sup>Ab</sup>	10.64 <sup>Aa</sup>
T3	5.91 <sup>Bd</sup>	6.06 <sup>Bd</sup>	6.63 <sup>Bc</sup>	8.57 <sup>Bb</sup>	9.23 <sup>Ba</sup>
T4	6.10 <sup>Bd</sup>	6.15 <sup>Bd</sup>	6.81 <sup>Bc</sup>	8.74 <sup>Bb</sup>	9.43 <sup>Ba</sup>
<b>Acetaldehyde (µml/100 g)</b>					
C	263.41 <sup>Ba</sup>	243.70 <sup>Cb</sup>	193.38 <sup>Cc</sup>	154.89 <sup>Cd</sup>	112.71 <sup>Ce</sup>
T1	261.12 <sup>Ba</sup>	245.67 <sup>Ccb</sup>	193.67 <sup>Cc</sup>	158.12 <sup>Cd</sup>	117.31 <sup>Ce</sup>
T2	270.32 <sup>Ba</sup>	249.70 <sup>CDab</sup>	201.26 <sup>Cc</sup>	162.86 <sup>Cd</sup>	121.12 <sup>Ce</sup>
T3	318.67 <sup>Aa</sup>	278.42 <sup>Bb</sup>	235.16 <sup>Bc</sup>	207.86 <sup>Bd</sup>	168.11 <sup>Be</sup>
T4	331.35 <sup>Aa</sup>	308.43 <sup>Ab</sup>	259.53 <sup>Ac</sup>	234.08 <sup>Ad</sup>	178.71 <sup>Ae</sup>
<b>Diacetyl (µml/100 g)</b>					
C	17.50 <sup>Ca</sup>	19.81 <sup>Ca</sup>	17.15 <sup>Ca</sup>	11.73 <sup>Cb</sup>	9.31 <sup>Cc</sup>
T1	17.61 <sup>Ca</sup>	19.71 <sup>Ca</sup>	17.65 <sup>Ca</sup>	11.89 <sup>Cb</sup>	9.51 <sup>Cc</sup>
T2	18.02 <sup>Ca</sup>	19.90 <sup>Ca</sup>	17.75 <sup>Ca</sup>	11.90 <sup>Cb</sup>	9.89 <sup>Cc</sup>

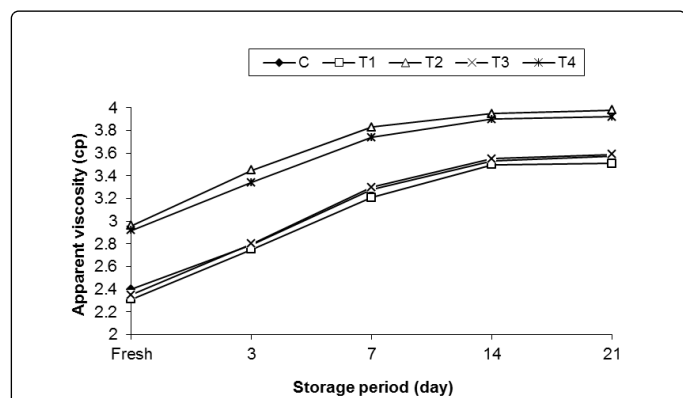
T3	21.88 <sup>Ba</sup>	22.52 <sup>Ba</sup>	21.76 <sup>Ba</sup>	19.86 <sup>Bb</sup>	15.46 <sup>Bc</sup>
T4	24.76 <sup>Aa</sup>	25.64 <sup>Aa</sup>	23.39 <sup>Aa</sup>	21.62 <sup>Ab</sup>	17.67 <sup>Ac</sup>

C: yoghurt made from liquid skim milk without maltodextrin with 3% yoghurt starter culture  
T1: yoghurt made from liquid skim milk without maltodextrin inoculated with 1.5% yoghurt culture+1.5% *Lb. acidophilus*  
T2: yoghurt made from liquid skim milk with 2% maltodextrin inoculated with 1.5% yoghurt culture+1.5% *Lb. acidophilus*  
T3: yoghurt made from liquid skim milk without maltodextrin inoculated with 1.5% yoghurt culture+1% *Bifi. bifidum*  
T4: yoghurt made from liquid skim milk with 2% maltodextrin inoculated with 1.5% yoghurt culture+1.5% *Bifi. bifidum*  
A, B, C: Means with same letter among treatments in the same storage period are not significantly different.  
a, b, c : Means with same letter for same treatment during storage periods are not significantly different.

**Table 1:** Chemical compositions of low fat bio-yoghurt containing different probiotic strains and traditional culture as well as 2% maltodextrin along the storage at 4°C for 21 days.

### Apparent viscosity

As shown in Figure 1, it was illustrated that adding a 2% maltodextrin to the yoghurt formula caused a significant increase in apparent viscosity of low-fat yoghurt products. On the other hand, the type of starter culture had no significant effect on the viscosity values in bio-yoghurt product. The higher viscosity of low fat yoghurt containing maltodextrin could be attributed to entrapment of water in the gel due to the combined effect of gelation of maltodextrin and milk. Bisar et al. [10] suggested that, the increase in apparent viscosity in skimmed fermented dairy product fortified with maltodextrin could be attributed to partial hydrolysis of starch from a variety of initiation and acceleration of maltodextrin gel formation. The outerlinear chains of amylopectin are thought to interact with amylose, thus reducing their self-association, and leading to the formation of a hydrated common network. Our results agree with Oliveira et al. [9], who reported that maltodextrins are maltooligosaccharides with a degree of polymerization ranging from three to nine and often act as fat replacers and bulking agents in dairy product. It is clear that, the apparent viscosity of all low-fat yoghurt products gradually increased during cold storage period. These results are confirmed with the data obtained by Donkor et al. [43] who noticed that, the viscosity values of different probiotic yoghurts increased during the storage at 5°C.



**Figure 1:** Apparent viscosity values (at  $\gamma=145.8 \text{ s}^{-1}$ ) of low fat bio-yoghurt containing different probiotic strains and traditional culture as well as 2% maltodextrin along the storage at 4°C for 21 days. \*See Table 1 for details.

### Microbiological analysis

As shown in Table 2, it could be noticed that, the counts of *Str. thermophilus* and *Lb. delbrueckii ssp. bulgaricus* were higher in traditional yoghurt sample made with 3% yoghurt culture than their counts in all other treatments. *Str. thermophilus* and *Lb. delbrueckii ssp. bulgaricus* counts slightly decreased during the first week of storage and then gradually decreased till the end of the storage period. Same findings were obtained by El Batawy [24] who found the same decrease trend in the growth of yoghurt cultures during cold storage period. In the beginning of storage period, the counts of *Lb. acidophilus* in T1 and T3 bio-yoghurt were 7.88 and 7.81 log cfu/ml respectively. At the end of storage period, the counts decreased to 6.46 and 6.52 log cfu/ml, in order. In the beginning of storage period, *Bifidobacterium ssp.* counts were 7.12 and 7.94 log cfu/ml in T3 and T4 fresh bio-yoghurt respectively. *Bifidobacterium ssp.* counts gradually decreased as the storage period progressed to be 5.72 and 6.62 log cfu/ml in T3 and T4 bio-yoghurt, respectively at the end of the storage period. From the results, it could be report that, gradual decrease were observed in the viability of probiotic strains (*Lb. acidophilus* and *Bifidobacterium ssp.*) in bio-yoghurt during the cold storage period. The gradual decrease in probiotic strains counts was due to the sensitivity of this bacteria to acid development along the storage period. Our results are in harmony with those obtained by Ibrahim et al.; Oliveira et al.; Paseephol and Sherkat; Bisar et al. [9,10,44,45] who reported that the *lactobacillus* and *bifidobacteria* growth declined in their viability during storage.

From these results, it could be observed that the addition of 2% maltodextrin as prebiotic in the manufacture of bio-yoghurt had no significant effect on the viability of yoghurt cultures (*Str. thermophilus* and *Lb. delbrueckii ssp. bulgaricus*) and *Lb. acidophilus* strains. This may be due to the yoghurt culture and different *lactobacilli* are unable to efficiently utilize maltodextrin during the growth [46]. On the other hand, fortification bio-yoghurt with maltodextrin had significantly enhanced the viability of *Bifid. bifidum* probiotic strain along the storage period. The growth enhancement of *Bifidobacterium ssp.* in the presence of maltodextrin was probably due to the ability of this strain to produce the enzyme that hydrolyses maltodextrin to glucose for growth [30]. These results are in agreement with the observations of Bisar et al. [10] who concluded that, maltodextrin was found to have good results in stimulation the probiotic bacterial count.

Treatments	Storage period (day)				
	Fresh	3	7	14	21
<i>Str. Thermophilus</i>					
C	7.91 <sup>Aa</sup>	7.67 <sup>ABa</sup>	7.26 <sup>Aa</sup>	6.86 <sup>Ab</sup>	6.59 <sup>Ac</sup>
T1	7.40 <sup>Ba</sup>	7.31 <sup>Ba</sup>	6.86 <sup>Ba</sup>	6.64 <sup>Bb</sup>	6.47 <sup>Ab</sup>
T2	7.38 <sup>Ba</sup>	7.32 <sup>Ba</sup>	6.85 <sup>Ba</sup>	6.62 <sup>Ba</sup>	6.47 <sup>Aa</sup>
T3	7.37 <sup>Ba</sup>	7.31 <sup>Ba</sup>	6.89 <sup>Ba</sup>	6.68 <sup>Bb</sup>	6.40 <sup>Ab</sup>
T4	7.35 <sup>Ba</sup>	7.28 <sup>Ba</sup>	6.81 <sup>Ba</sup>	6.70 <sup>Bb</sup>	6.44 <sup>Ac</sup>
<i>Lb. delbrueckii ssp. Bulgaricus</i>					
C	8.25 <sup>Aa</sup>	8.14 <sup>Aa</sup>	8.00 <sup>Aa</sup>	7.42 <sup>Ab</sup>	6.65 <sup>Ac</sup>
T1	7.63 <sup>Ba</sup>	7.52 <sup>Ba</sup>	7.11 <sup>Bab</sup>	6.81 <sup>Bc</sup>	6.32 <sup>Bd</sup>
T2	7.61 <sup>Ba</sup>	7.45 <sup>Ba</sup>	6.98 <sup>Bb</sup>	6.82 <sup>Bc</sup>	6.21 <sup>Bd</sup>
T3	7.55 <sup>Ba</sup>	7.46 <sup>Ba</sup>	7.13 <sup>Bb</sup>	6.75 <sup>Bc</sup>	6.14 <sup>Bd</sup>
T4	7.62 <sup>Ba</sup>	7.35 <sup>Ba</sup>	7.04 <sup>Bb</sup>	6.50 <sup>Bc</sup>	6.11 <sup>Bd</sup>
<i>Lb. Acidophilus</i>					
T1	7.88 <sup>Aa</sup>	7.73 <sup>Aa</sup>	7.56 <sup>Aab</sup>	7.05 <sup>Ab</sup>	6.46 <sup>Ac</sup>
T3	7.81 <sup>Aa</sup>	7.70 <sup>Aa</sup>	7.61 <sup>Aa</sup>	7.11 <sup>Ab</sup>	6.52 <sup>Ac</sup>
<i>Bifidobacterium ssp.</i>					
T2	7.12 <sup>Ba</sup>	7.10 <sup>Bab</sup>	6.81 <sup>Bb</sup>	6.03 <sup>Bc</sup>	5.72 <sup>Bd</sup>
T4	7.94 <sup>Aa</sup>	7.63 <sup>Aa</sup>	7.13 <sup>Aab</sup>	6.83 <sup>Ab</sup>	6.62 <sup>Ac</sup>
<i>Yeast and mould</i>					
C	ND	ND	2.31	2.51	3.77 <sup>A</sup>
T1	ND	ND	2	2.4	2.58 <sup>B</sup>
T2	ND	ND	2	2.31	2.45
T3	ND	ND	ND	2	2.75
T4	ND	ND	ND	2	2.48
<p>C: yoghurt made from liquid skim milk without maltodextrin with 3% yoghurt starter culture                      T1: yoghurt made from liquid skim milk without maltodextrin inoculated with 1.5% yoghurt culture+1.5% <i>Lb. acidophilus</i>                      T2: yoghurt made from liquid skim milk with 2% maltodextrin inoculated with 1.5% yoghurt culture+1.5% <i>Lb. acidophilus</i>                      T3: yoghurt made from liquid skim milk without maltodextrin inoculated with 1.5% yoghurt culture+1% <i>Bifi. bifidium</i>                      T4: yoghurt made from liquid skim milk with 2% maltodextrin inoculated with 1.5% yoghurt culture+1.5% <i>Bifi. bifidium</i>                      A, B, C: Means with same letter among treatments in the same storage period are not significantly different.                      a, b, c : Means with same letter for same treatment during storage periods are not significantly different.                      ND: not detected in samples.</p>					

**Table 2:** Bacteriological properties (log cfu/ml) of low fat bio-yoghurt containing different probiotic strains and traditional culture as well as 2% maltodextrin along the storage at 4°C for 21 days.

In general, the food industry has targeted populations over  $10^6$  probiotics/g at the time of consumption of strain added to food [47]. However it is clear that, the viability of probiotic strains (*Lb. acidophilus* and *Bifidobacterium ssp.*) in bio-yoghurt made with or

without maltodextrin as prebiotic were higher during the storage at (4°C for 14 days) than the recommended minimum levels (106 cfu/ml or g). while, the viability of the same strains bio-yoghurt made with maltodextrin as prebiotic were higher during the storage at (4°C for 21

days) than the recommended minimum levels ( $10^6$  cfu/ml or g). Tabatabaie and Mortazavi [48] reported that, *Bifidobacterium bifidum* were found to be extremely stable in bio-yoghurt during the 5 weeks storage period and survives slightly better in the presence of prebiotic. Hekmat; FAO/WHO and Salem et al. [49-51] reported that the standard for any food sold with health claims from the addition of probiotics that it must contain at least  $10^6$ - $10^7$  cfu/gram or ml of viable probiotic bacteria.

Generally, yeast and mould counts could not be detected in all fresh and 3 days refrigerated stored samples. Moreover, yeast and mould

counts could be observed and counted after 7 days of the storage in treatment samples (C, T1 and T2), and after 14 days in treatments (T3 and T4). These counts slightly increased as the storage period progressed and this is may be due to the post contamination in these samples after manufacturing and during filling the products [24]. All fresh and 21 days refrigerated stored bio-yoghurts containing different probiotic and traditional cultures as well as inulin were free from coliform bacteria.

Characteristics	Storage period (day)					
	Treatments	Fresh	3	7	14	21
Appearance	C	8 <sup>Aa</sup>	9 <sup>Aa</sup>	7 <sup>Aab</sup>	6 <sup>Ab</sup>	6 <sup>Ab</sup>
	T1	9 <sup>Aa</sup>	9 <sup>Aa</sup>	8 <sup>Aa</sup>	7 <sup>Ab</sup>	6 <sup>Ab</sup>
	T2	9 <sup>Aa</sup>	8 <sup>Aa</sup>	8 <sup>Aa</sup>	6 <sup>Ab</sup>	6 <sup>Ab</sup>
	T3	8 <sup>Aa</sup>	9 <sup>Aa</sup>	8 <sup>Aa</sup>	7 <sup>Ab</sup>	6 <sup>Ac</sup>
	T4	8 <sup>Aa</sup>	9 <sup>Aa</sup>	8 <sup>Aa</sup>	6 <sup>Ab</sup>	6 <sup>Ab</sup>
Body and Texture (30)	C	24 <sup>Ca</sup>	24 <sup>Ca</sup>	23 <sup>Cab</sup>	20 <sup>Bbc</sup>	19 <sup>Cc</sup>
	T1	26 <sup>Ba</sup>	26 <sup>Ba</sup>	25 <sup>Bab</sup>	24 <sup>Bab</sup>	23 <sup>Bb</sup>
	T2	29 <sup>Aa</sup>	28 <sup>Aa</sup>	28 <sup>Aab</sup>	27 <sup>Ab</sup>	25 <sup>Ac</sup>
	T3	26 <sup>Ba</sup>	25 <sup>Ba</sup>	25 <sup>Ba</sup>	24 <sup>Bb</sup>	22 <sup>Bc</sup>
	T4	29 <sup>Aa</sup>	28 <sup>Aa</sup>	28 <sup>Aa</sup>	27 <sup>Aa</sup>	25 <sup>Ab</sup>
Flavour (60)	C	51 <sup>Ca</sup>	50 <sup>Ca</sup>	48 <sup>Cb</sup>	40 <sup>Cc</sup>	33 <sup>Cd</sup>
	T1	55 <sup>Ba</sup>	55 <sup>Ba</sup>	53 <sup>Bb</sup>	45 <sup>Bc</sup>	42 <sup>Bd</sup>
	T2	58 <sup>Aa</sup>	58 <sup>Aa</sup>	56 <sup>Aa</sup>	49 <sup>Ab</sup>	45 <sup>Ac</sup>
	T3	56 <sup>Ba</sup>	56 <sup>Ba</sup>	53 <sup>Bb</sup>	46 <sup>Bc</sup>	42 <sup>Bd</sup>
	T4	58 <sup>Aa</sup>	58 <sup>Aa</sup>	57 <sup>Aa</sup>	51 <sup>Ab</sup>	45 <sup>Ac</sup>
Total (100)	C	83 <sup>Ca</sup>	83 <sup>Ca</sup>	78 <sup>Cb</sup>	66 <sup>Cc</sup>	58 <sup>Cd</sup>
	T1	90 <sup>Ba</sup>	90 <sup>Ba</sup>	86 <sup>Bb</sup>	76 <sup>Bc</sup>	71 <sup>Bd</sup>
	T2	96 <sup>Aa</sup>	94 <sup>Aa</sup>	92 <sup>Aa</sup>	82 <sup>Ab</sup>	76 <sup>Ac</sup>
	T3	90 <sup>Ba</sup>	90 <sup>Ba</sup>	86 <sup>Bb</sup>	77 <sup>Bc</sup>	70 <sup>Bd</sup>
	T4	95 <sup>Aa</sup>	95 <sup>Aa</sup>	93 <sup>Aa</sup>	84 <sup>Ab</sup>	76 <sup>Ac</sup>

C: yoghurt made from liquid skim milk without maltodextrin with 3% yoghurt starter culture

T1: yoghurt made from liquid skim milk without maltodextrin inoculated with 1.5% yoghurt culture+1.5% *Lb. acidophilus*

T2: yoghurt made from liquid skim milk with 2% maltodextrin inoculated with 1.5% yoghurt culture+1.5% *Lb. acidophilus*

T3: yoghurt made from liquid skim milk without maltodextrin inoculated with 1.5% yoghurt culture+1% *Bifi. bifidum*

T4: yoghurt made from liquid skim milk with 2% maltodextrin inoculated with 1.5% yoghurt culture+1.5% *Bifi. bifidum*

A, B, C: Means with same letter among treatments in the same storage period are not significantly different.

a, b, c : Means with same letter for same treatment during storage periods are not significantly different.

**Table 3:** Organoleptic properties of low fat bio-yoghurt containing different probiotic strains and traditional culture as well as 2% maltodextrin along the storage at 4°C for 21 days.

This may be due to the efficient heat treatment of the standardized milk (90°C for 10 min) and high sanitation conditions during manufacture and storage. In addition, the effect of acidity in different bio-fermented milk which plays an important role in reduction of the growth rate of coliform bacteria. These results are in accordance with those reported by El-Nagar and Shenan; El Batawy [23,24].

### Organoleptic properties

To produce healthy fermented milk products, it must be firstly organoleptically acceptable. Therefore, organoleptic properties of bio-yoghurt fermented with different probiotic and traditional cultures as well as maltodextrin were evaluated along the storage at 4°C for 21 days. As shown in Table 3, there were slight differences in appearance scores among all treatments during the first 7 days of storage period. After that, the appearance score points decreased as the storage period progressed reaching 6 points for all samples at the end of storage period (21 days). It is clear that, the type of starter culture used and addition of maltodextrin did not affect on the appearance score in different bio-yoghurt.

The panel found significant differences for each sample for flavour, body and texture and overall acceptability, which reflects the advantages of probiotic culture and maltodextrin as effective components on the general sensory properties of yogurt. The data indicates that, the highest of body and texture scores (29) were recorded for fresh bio-yoghurt fermented with 1.5% yoghurt culture +1.5% *Bifi. Bifidum* and fortified with 2% maltodextrin, (T4). While, fresh control yogurt made with 3% yoghurt culture (C) reached the least body and texture scores (24) compared with all other samples. The highest remarkable flavour score points (58) were in bio-yoghurt milk fermented with yoghurt and *Bifi. bifidum* cultures with or without maltodextrin (T2 and T4), followed by (T2) bio-yoghurt containing *Lb. acidophilus*. The data indicated that, inoculation of *Bifi. Bifidum* and *Lb. acidophilus* as starter culture in bio-yoghurt production improves the flavour scores. From these data, it could be stated that, the type of starter culture used had significantly effect on the flavour and body and texture scores in different bio-yoghurt.

It could be observed that, flavour, body and texture and total score points of bio-yoghurt were enhanced by adding the 2% maltodextrin as prebiotic substance. This provides that, maltodextrin plays a key role in enhancing the test and texture of bio-yoghurt. The increased body and texture scores of maltodextrin containing bio-yoghurt could be due to the water binding capacity of low molecular weight polymers (dextrans) present in maltodextrin [52]. Therefore, it could be reported that the sensory response to the yoghurt samples demonstrated that the use of probiotic culture combination and maltodextrin positively influenced the overall sensory characteristics. These results agree with Ranjeeta [53] who showed that maltodextrin improved the body and texture score ( $p < 0.05$ ) of some dairy products compared with other bulking agents batches. Maltodextrins are frequently complex mixtures of molecular species ranging from glucose to long polymeric (linear and branched) chains [54]. Saccharides, oligosaccharides and polysaccharides form complexes with proteins and lipids which are known to contribute to the texture of food stuffs. Moreover Hyvonen et al. [55] stated that some polysaccharides such as maltodextrin could be used as bodying agents in the fat-free ice creams significantly increase flavor release, fattiness, creaminess and melting rate of the ice cream. Also, Bisar et al. [10] found that, maltodextrin could be enhance the sensory properties of fermented milk compared with inulin.

The total score points of all yoghurt samples were higher till 3rd of the storage period, followed by gradual decrease till the end of storage period. This decrease may be due to the acidity development or the production of other microbial exerted metabolism which affect on the rheological and sensory properties. The results also indicated that, the shelf life of bio-yoghurt could be extended more than 14 days of the storage at 4°C because there was a drop in organoleptic properties of all samples. These findings are in agreement with results reported by Abo Iaina; El Batawy [24,56].

### Conclusion

Finally, it could be concluded that, different probiotic strains such as *Lb. acidophilus* and *Bifi. bifidum* could be combined with yoghurt culture for produce low-fat bio yoghurt. Addition of maltodextrin during low fat bio-yoghurt manufacture could be enhancing the viability of probiotic strains and sensory properties of final product during the storage period, and that is recommended by this work to use it in industry.

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