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Effect of Adding Bifidobacteria and Turmeric on the Quality of Yoghurt

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Abstract :

Effect of adding bifidobacteria and turmeric, on the quality of yoghurt monitors the survival of different strains of bifidobacteria and changes of chemical, microbiological and organoleptic properties during storage of yoghurt were investigated. Results indicated that yoghurt treatments which made with normal starter had higher acidity than those of corresponding treatments made with bifidobacteria strains. Titratable acidity decreased as the amount of *B. bifidum* ATCC 15696, *B. bifidum* ATCC 29521 and *B. longum*BL-04increased. Samples which made with incorporating bifidobacteria strains have a higher pH values than control sample. There was a positive correlation between the amount added of bifidobacteria and pH. Neither the strain nor the species of bifidobacteria significantly affected ($p \leq 0.05$) the total solids content of yoghurt. Diacetyl methyl carbinol content of all set yoghurt treatments increased gradually up to the sixth day of storage, then decreased as storage period progressed. No significant differences recorded among yoghurt treatments in total protein and fat content.Total bacterial counts of all yoghurt treatments increased up to the third day of storage then decreased up to the end of storage period. Incorporating of bifidobacteriacaused a significant decrease of the counts of moulds and yeasts. Even after 9 days of cold storage, the counts of bifidobacteria in all yoghurt treatments were still higher than the minimum level ($10^5 - 10^6 / ml$) that should be present at the consumption of the product to achieve the beneficial effect of bifidobacteria. Probiotic yoghurt treatments which made by adding *B. bifidum* ATCC 15696 were not

significantly different ($p > 0.05$) of organoleptic properties from corresponding treatments made by adding either *B. bifidum* ATCC 29521 or *B. longum* BL-04. After 12 days of cold storage, the count of bifidobacteria with turmeric was still higher than the minimum level ($10^5 - 10^6 / \text{ml}$) that should be present at the consumption of the product. The turmeric concentration did not affect on the survival of bifidobacteria strains. Yoghurt treatments which made by adding bifidobacteria with turmeric slightly decreased organoleptic properties.

Key words: Bifidobacteria strain, Turmeric, Yoghurt quality.

Introduction

Yoghurt is the most popular fermented milk in Egypt and all over the world. It has a sharp refreshing acid taste and the typical flavour described as being similar to walnuts.

Yoghurt is one of the best-known of the foods that contain probiotics. Yoghurt is defined by the Codex Alimentarius of 1992 as a coagulated milk product that results from the fermentation of lactic acid in milk by *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus* (**Chandan and Shasant, 1993**).

The consumption of yoghurt has been increased in Egypt and world because of using the pure culture & applying of modern equipment which resulted in continuous processes introducing wide range of flavoured yoghurt supplementing the yoghurt flora with *L. acidophilus* for the purpose of increasing the dietetic value (**Rastic and Kurmann, 1978**).

Hoolihan (2001) mentioned that yoghurt and milk to which probiotic bacteria have been added and fermented milk products are the primary food sources of probiotics. The synergistic effect exists between components in dairy foods and probiotic cultures and that there are components in milk that turn on the beneficial genes in probiotic bacteria making dairy foods an excellent vehicle for introducing these bacteria into the gut.

Marshall et al., (1988) reported that the value of yoghurt in human nutrition is based not only on the strict nutritive effect of milk from which it is made and the chemical changes of milk constituents occurring during lactic acid fermentation but also on the beneficial effect to intestinal microflora particularly in certain conditions and prophylactic and healing effects. The fine cured particles formed of yoghurt is more easily digested by enzymes than large casein particles of ordinary milk being formed by gastric juice, also the presence of lactic acid and culture microflora results in a significant hydrolysis of proteins. Fat in yoghurt is easier to digest than fat in the ordinary milk. Yoghurt has a positive healing effect when antibiotic and radiation therapy

applied and for people suffering from chronic constipation, diarrhea, colitis, intestinal intoxication, liver and bile disorders.

The value of yoghurt in human nutrition is based not only on the strict nutritive effect of milk from which it is made and the chemical changes of milk constituents occurring during lactic acid fermentation, but also on the beneficial effect to intestinal microflora particularly in certain conditions and prophylactic and healing effects. Yoghurt has a positive healing effect when antibiotic and radiation therapy applied and for people suffering from chronic constipation diarrhea, colitis, intestinal intoxication, liver and bile disorders (**Guyonnetet al., 2007**).

The potential beneficial roles of bifidobacteria in the human intestine have been reported by **Kebaryet al., (2005)**. Bifidobacteria shows antagonistic effects towards enteropathogenic bacteria. *Lactobacilli* and bifidobacteria break down carcinogenic N-nitrosamines and also suppress liver tumorigenesis in mice. The dietary administration of *Bif. Bifidum* in patients reduced ammonia free serum phenol and free amino nitrogen in blood.

Minamiet al., (2015) reported that that *bifidobacteriade* decreased the level of cholesterol in rats alleviated of lactose intolerant individuals and synthesized many vitamins. Because of that many efforts have been devoted to incorporate the bifidobacteria in dairy products such as fermented milks, ferment-milk beverages, butter milk sour cream, fresh cheese, baby foods, as well as pharmaceutical preparations and also livestock feed supplements.

Wickenberg (2015) reported that turmeric is a well-known remedy used in ancient Indian traditional medicine and cosmetics. It serves as a multipurpose herbal remedy for practitioners of Ayurveda, Unani and practitioners of traditional Chinese medicine. Turmeric is also used to treat asthma dysmenorrhoea (painful menstruation), psoriasis (an inflammatory skin disease), eczema, arthritis and hepatic and digestive disorders, and to prevent and treat cardiovascular diseases. The therapeutically components of turmeric are thought to be its polyphenolic compounds including curcuma oil (particularly dl-ar-turmerone) and various curcuminoids especially curcumin which exhibits a wide range of biological activities.

Khandelwalet al., (2006) reported that turmeric contains various chemical constituents such as a-tumerone, b-tumerone, zingiberine and

curcumin. The therapeutical components of turmeric are thought to be its polyphenolic compounds including curcuma oil (particularly dl-ar-turmerone) and various curcuminoids especially curcumin which exhibits a wide range of biological activities.

The objectives of this study were to investigate the possibility of making a good quality probiotic yoghurt by replacing the normal yoghurt starter with different strains of bifidobacteria, investigate the effect of adding turmeric on the survival of bifidobacteria and the quality of yoghurt, monitor the survival of bifidobacteria and changes of chemical, microbiological, and organoleptic properties during storage of yoghurt.

Materials And Methods

Materials:

Raw milk:

Fresh bulk buffaloe's milk was obtained from the herd of Faculty of Agriculture, Menoufia University, Shibin El-kom, Egypt.

Turmeric:

Turmeric as powder was obtained from herbalist, at shebin El-Kom City, Menoufia Governorate.

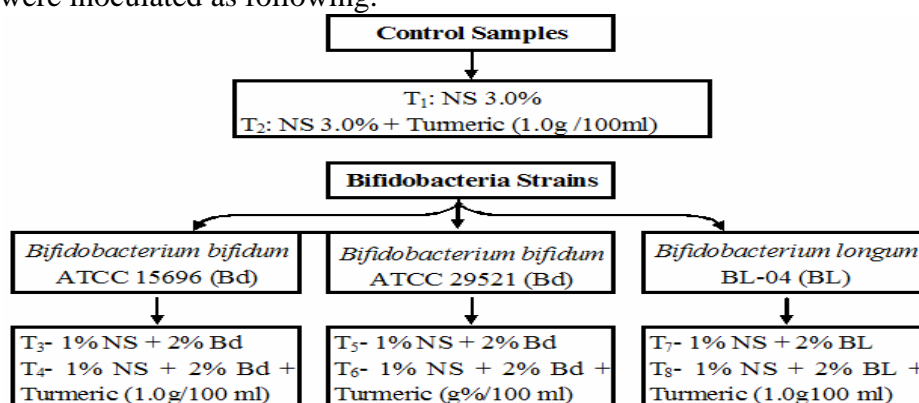
Bacterial strains and propagation:

Active *Streptococcus thermophilus* EMCC 1043 and *Lactobacillus delbrueckii sub sp. bulgaricus* EMCC 1102 were obtained from Cairo Mircen, Ain Shams University, Egypt. *Bifidobacterium bifidum* ATCC 15696 and *Bifidobacterium bifidum* ATCC 29521, were gratefully obtained from Dr. Linda J. Brady's Lab (Department of Food Science and Nutrition, University of Minnesota, USA), while *Bifidobacterium longum* BL-04 was obtained from Rodia, Madison, WI. USA. Bifidobacteria strains were activated individually by three successive transfers in modified MRS (Ventling and Mistry, 1993) followed by three successive transfers in sterile 10% reconstituted non-fat dry milk, and incubated at 37°C under anaerobic condition. *Lactobacillus bulgaricus* and *Streptococcus thermophilus* were activated individually by three successive transfers in sterile 10% reconstituted non-fat dry milk.

Methods:

Manufacture of yoghurt:

Buffaloe's milk was standardized to 5% fat. Milk was heated to 85°C for 20 min then cooled to 42°C. Milk was divided to ten portions. The first portion (control T₁) was inoculated by 3% of the normal starter (NS) (1.5% Streptococci + 1.5% Lactobacilli). The other nine portions were inoculated as following:



The inoculated batches were packed in plastic cups and incubated at 42°C until coagulation. All yoghurt treatments were stored in the refrigerator ($\pm 6^\circ\text{C}$) for 9 days and were sampled when fresh and at 3, 6 and 9 days for microbiological, chemical, rheological and sensory evaluation.

In the second part of this study it was concerned to study the effect of adding turmeric on survival of *bifidobacteria* and sensory evaluation of yoghurt. Preliminary experiment was carried out to choose the best amount of turmeric. 5 yoghurt treatments were made from 5.0% buffalo's milk with adding turmeric at rate of 0.0, 1.0, 2.0, 3.0 and 4.0 g/100 ml milk. The obtained results revealed that yoghurt made with 1.0 g/100 ml milk was the most acceptable yoghurt. Eight yoghurt treatments were made from 5.0% buffalo's milk. Four of them were made without adding turmeric, but control yoghurt was made by inoculating 3.0% normal starter (NS). To the other three treatments 1.0% normal starter and 2.0% *Bifidobacterium bifidum* ATCC15696, *Bifidobacterium bifidum* ATCC 29521 and *Bifidobacterium longum* BL-04 were added individually. The other four yoghurt treatments were made as described above except that turmeric was added at the rate of 1.0 g/100 ml milk to all milk treatments. All yoghurt treatments were made, stored for 12 days and sampled at 1, 3, 6, 9 and 12 days for sensory evaluation and counting bifidobacteria. The experiment was triplicates.

Microbiological analysis:

The total bacterial count was determined using standard plate count agar (**Marth, 1978**). Yeasts and moulds were enumerated on Potato Dextrose Agar (acidified) medium (**Difco, 1953**). Modified MRS agar was used for enumerating Bifidobacteria (**Ventling and Mistry, 1993**). To each 100 ml of modified MRS. 5ml of the solution was added before pouring plates (**Samona and Robinson, 1991**):

Analytical Methods:

pH value:

pH value was determined according to **Ling (1963)**, the pH value was measured using pH meter (Jenway LTD, FelstedDunmow, Essex UK).

Determination of acidity:

Acidity was determined as lactic acid percent according to **Ling (1963)**.

Total solid (TS):

Total solid was determined according to the Official Method (**A.O.A.C, 1990**).

Fat content:

Fat content was determined by original Gerber's method according to **Ling (1963)**.

3.2.3.6. Total protein content:

Total protein was determined according to the Official Method (**A.O.A.C, 1990**).

Determination of diacetyl (DA) and acetyl methyl carbinol (AMC) content:

DA + AMC were determined according to the method of **Brandel (1960)**.

Sensory evaluation:

Yoghurt was assessed according to **Keব্য and Hussein (1999)** by ten panelists from the Staff of Dairy Science and Technology Department, and Food Science and Technology Department, Faculty of Agriculture, Menoufia University. Using the following score points: flavor (45 points), acidity (10 points), body and texture (35 points) and appearance (10 points).

Statistical Analysis:

Statistical analysis were performed by using computer program statistical package for social science (SPSS), and compared with each other using the suitable test. All obtained results were tabulated. Statistical analysis has been achieved using IMB-P-C computer by SPSS, program **SPSS (1998)**.

Results And Discussion

Part 1: Effect of incorporating bifidobacteria on the quality of probiotic yoghurt:

Chemical properties of yoghurt:

Titratable acidity:

Changes in titratable acidity of set yoghurt made with bifidobacteria (*B. bifidum* ATCC 15696, *B. bifidum* ATCC 29521 and *B. longum* BL-04) during the storage period are present in Table (1). It could be observed that there were significant differences among set yoghurt treatments ($p \leq 0.05$). Yoghurt treatments which made with normal starter had higher acidity than those of corresponding treatments made with bifidobacteria strains. Titratable acidity decreased as the amount of *B. bifidum* ATCC 15696, *B. bifidum* ATCC 29521 and *B. longum* BL-04 increased which might be due to the lower acid production of bifidobacteria strains and / or their antagonistic effect on the other bacteria (**Kebaryet al., 2007**). Treatments which made with adding *B. bifidum* ATCC 29521 had higher acidity than those of corresponding yoghurt treatments which made with either *B. bifidum* ATCC 15696 or *B. longum* BL-04. These results may be due to the differences in acid production, which is strain and species dependant (**Takahashi et al., 2004**). On the otherhand, titratable acidity of all set yoghurt treatments increased as storage period advanced. Set yoghurt samples at 9 days of storage had the highest titratable acidity and were significantly ($p \leq 0.05$) different from samples at 6, 3 and 1 time of storage. Titratable acidity of T₁, T₂, T₃, T₄, T₅, T₆, T₇, T₈, T₉ and T₁₀ were 0.98, 0.87, 0.82, 0.78, 0.90, 0.86, 0.81, 0.87, 0.81 and 0.77%, respectively, when were fresh, but the corresponding values of titratable acidity at 9 days of storage were 1.16, 1.06, 0.97, 0.94, 1.09, 1.05, 0.98, 1.06, 0.98 and 0.95% successively. These results are in agreement with those reported by **Badran(2004)**.

PH values:

Table (1) also; show the changes in pH values of yoghurt samples. pH values of all yoghurt treatments followed contradictory trends of titratable acidity. It could be observed that samples made with incorporating bifidobacteria strains have higher pH values than control sample. There was a positive correlation between the amount added bifidobacteria and pH. pH values increased as the amount of *B. bifidum* ATCC 15696, *B. bifidum* ATCC 29521 and *B. longum* BL-04 increased. On the other hand, yoghurt treatments those made with incorporating *B. bifidum* ATCC 29521 exhibited a lower pH values than those of corresponding treatments made with incorporating *B. bifidum* ATCC 15696. These results might be attributed to the low acid production of bifidobacteria and the production of acid is strain dependant as explained previously. pH values of all yoghurt treatments decreased throughout the storage period, which might be due to the availability of lactose and development of acidity. Samples at 9 days old had the lowest pH value and were significantly ($p \leq 0.05$) different from the samples at any storage period see Table (9&10). pH values of fresh yoghurt T₁, T₂, T₃, T₄, T₅, T₆, T₇, T₈, T₉ and T₁₀ were 4.75, 4.60, 4.68, 4.71, 4.58, 4.62, 4.66, 4.62, 4.64 and 4.69%, respectively. The corresponding pH values at the end of storage were 4.26, 4.37, 4.43, 4.46, 4.31, 4.36, 4.41, 4.35, 4.41 and 4.43% in the same order. These results are in agreement with those reported by **Badawi (2004)**.

Total solids content:

Total solids content of set yoghurt samples are present in Table (2). Yoghurt treatments were not significantly ($p > 0.05$) different from each other. These results mean that neither the strain nor the species of bifidobacteria affected significantly ($p \leq 0.05$) the total solids content of yoghurt (**Kebaryet al., 2008**). Also, there is no significant effect ($p > 0.05$) of bifidobacteria concentrations on total solids content. Total solids contents of all set yoghurt treatments did not change significantly ($p > 0.05$) as storage period progressed. Total solids contents were 15.69, 15.66, 15.71, 15.67, 15.68, 15.66, 15.69, 15.69, 15.67 and 15.72% when fresh for treatments T₁, T₂, T₃, T₄, T₅, T₆, T₇, T₈, T₉ and T₁₀, respectively. While it reached to 15.72, 15.68, 15.66, 15.67, 15.67, 15.66, 15.70, 15.71, 15.68 and 15.69% at the 9th day of storage period, in the same order. These results are in accordance with the results obtained by **Pradyumanand Mishra (2004)**.

Diacetyl and acetyl methyl carbinol (DA + AMC):

The effect of bifidobacteria (*B. bifidum* ATCC 15696, *B. bifidum* ATCC 29521 and *B. longum* BL-04) on diacetyl and acetyl methyl carbinol ($\mu\text{g} / 100 \text{ g}$) (DA + AMC) during the storage period of set yoghurt treatments is shown in Tables (2). Diacetyl and acetyl methyl carbinol content of fresh treatments T₁, T₂, T₃, T₄, T₅, T₆, T₇, T₈, T₉ and T₁₀ were 38.11, 42.62, 43.78, 45.36, 41.32, 42.71, 44.43, 42.65, 44.21 and 46.71 $\mu\text{g} / 100 \text{ g}$, respectively, while their values at sixth day of storage were 68.30, 62.03, 69.14, 74.97, 66.35, 70.51, 76.45, 68.12, 72.14 and 77.85 $\mu\text{g} / 100 \text{ g}$, successively. The corresponding diacetyl methyl carbinol content at the 9th day of storage period were 49.89, 50.21, 55.37, 61.34, 53.04, 63.13, 66.22, 54.35, 60.47 and 68.32 $\mu\text{g}/100 \text{ g}$, in the same order. These results indicated that diacetyl methyl carbinol content of all set yoghurt treatments increased gradually up to the sixth day of storage, then decreased as storage period progressed. This decrease may be attributed to the ability of *Str. thermophilus* to reduce (DA + AMC) to acetone (Farag, 2002). Similar results were reported by El-Sonbaty et al., (2008) and Badawiet al., (2008). Incorporation of bifidobacteria caused a significant ($p \leq 0.05$) increase of DA + AMC content of set yoghurt and this increase was proportional to the amount added of bifidobacteria (Kebaryet al., 2008). On the other hand, treatments made with *B. bifidum* ATCC 29521 were not significantly ($p \geq 0.05$) different from corresponding treatments those made with adding either *B. bifidum* ATCC 15696 or *B. longum* BL-04, which means that neither the strains nor the species of bifidobacteria affect significantly ($p \leq 0.05$) the diacetyl and acetyl methyl carbinol content of set yoghurt.

Total protein content:

Data presented in Tables (3) show the protein content of set yoghurt made with bifidobacteria (*B. bifidum* ATCC 15696, *B. bifidum* ATCC 29521 and *B. longum* BL-04). Total protein content of set yoghurt treatments T₁, T₂, T₃, T₄, T₅, T₆, T₇, T₈, T₉ and T₁₀ were 5.56, 5.54, 5.52, 5.55, 5.53, 5.57, 5.55, 5.52, 5.51 and 5.52% respectively, when fresh, while the corresponding values at the end of storage period were 5.55, 5.50, 5.48, 5.52, 5.50, 5.54, 5.52, 5.49, 5.48 and 5.50% in the same order. There were no significant differences among yoghurt treatments in total protein (Table 3) which means that neither the addition of

B.bifidum ATCC 15696 or *B.bifidum* ATCC 29521 and *B. longum*BL-04 nor their concentrations affect significantly ($p > 0.05$) the total protein of set yoghurt. On the other hand, total protein content of all yoghurt treatments did not change significantly ($p > 0.05$) as storage period progressed. Similar trends were obtained by **Hussein and Kebary (1999) and Kebary et al., (2008)**.

Fat content:

The effect of bifidobacteria (*B.bifidum* ATCC 15696, *B.bifidum* ATCC 29521 and *B. longum*BL-04) on fat content of set yoghurt treatments is shown in Table (3). Fat content of set yoghurt treatments T₁, T₂, T₃, T₄, T₅, T₆, T₇, T₈, T₉ and T₁₀ were 5.3, 5.5, 5.5, 5.4, 5.5, 5.4, 5.4, 5.5, 5.5 and 5.3 respectively, when fresh, while the corresponding values at the end of storage period were 5.4, 5.5, 5.3, 5.5, 5.5, 5.4, 5.5, 5.3, 5.3 and 5.5 in the same order. There were no significant differences ($p > 0.05$) among set yoghurt treatments in fat content which means that either the addition of bifidobacteria (*B.bifidum* ATCC 15696, *B.bifidum* ATCC 29521 and *B. longum*BL-04) or their concentrations did not affect significantly ($p > 0.05$) the fat content of set yoghurt. On the other hand, fat content of all yoghurt treatments did not change significantly ($p > 0.05$) as storage period progressed (Tables 10, 19). The results are in agreement with those reported by **Kebary et al., (2008)**.

Microbiological properties:

Total bacterial count:

Data given in Table (4) show the total bacterial counts during the storage period of probiotic yoghurt made with bifidobacteria (*B. bifidum* ATCC 15696, *B. bifidum* ATCC 29521 and *B. longum*BL-04). Total bacterial counts (cfu $\times 10^7$ / ml) of fresh treatments T₁, T₂, T₃, T₄, T₅, T₆, T₇, T₈, T₉ and T₁₀ were 113, 98, 82, 75, 102, 85, 68, 91, 82 and 69 (cfu $\times 10^7$ / ml), respectively, while they reached their maximum counts at the third day of storage period and were 127, 109, 96, 83, 113, 96, 81, 102, 93 and 76 (cfu $\times 10^7$ / ml), successively. The corresponding counts at the end of storage period were 87, 65, 58, 41, 68, 56, 45, 63, 51 and 38 (cfu $\times 10^7$ / ml), in the same order. These results revealed that total bacterial counts of all yoghurt treatments increased up to the third day of storage then decreased up to the end of storage period. Similar trends were obtained by **Badawi (2004)**. On the other hand, total bacterial count decreased with the addition of bifidobacteria (*B. bifidum* ATCC 15696,

B. bifidum ATCC 29521 and *B. longum*BL-04). This decrease in total bacterial counts was proportional to the amount added of *B. bifidum* ATCC 15696, *B. bifidum* ATCC 29521 and *B. longum*BL-04 during the manufacture of probiotic yoghurt. These results may be due to the effect of antimicrobial substances produced by bifidobacteria which inhibited many gram positive and negative bacteria (**Vifayendra and Guypta, 1992**) and /or increasing the amount of bifidobacteria which cannot grow under aerobic condition. Probiotic yoghurt treatments those made by adding *B. bifidum* ATCC 15696 did not differ from corresponding treatments those made by adding either *B. bifidum* ATCC 29521 or *B. longum*BL-04 in total bacterial counts.

Moulds and yeasts:

Data presented in Table (4) show that yoghurt treatments were free from yeasts and moulds during the first three days of storage period. After that, they appeared towards the end of storage period. These results are in agreement with those reported by **Mehrizet al., (1993)** who found that, moulds and yeasts were only detected at the end of storage period. Also, appearance of yeasts and moulds after the 3rd day of storage period may be due to the post contamination. Incorporating of bifidobacteria caused a significant decrease of the counts of moulds and yeasts and this decrease was proportional to the amount added of bifidobacteria. This decrease might be due to the production of antimicrobial substance by bifidobacteria (**Kebary, 1995**). Incorporation of *B. bifidum* ATCC 15696, *B. bifidum* ATCC 29521 and *B. longum*BL-04 has the same effect to decrease the count of moulds and yeasts.

Bifidobacteria count:

Results in Table (5) indicated that the counts of bifidobacteria increased during the first three days of storage period then decreased as storage period advanced in all treatments (**Kebaryet al., 2007, Kebaryet al., 2008**). The count of bifidobacteria of fresh T₂, T₃, T₄, T₅, T₆, T₇, T₈, T₉ and T₁₀ were 173, 205, 223, 187, 213, 242, 156, 192 and 211 (CFU × 10⁵ / ml), respectively. Corresponding counts at the third day of storage period were 196,222, 256, 216, 245, 261, 177, 209 and 236 (CFU × 10⁵ / ml), in the same order, but they reached 113, 152, 176, 126, 168, 191, 109, 148 and 176 (CFU × 10⁵ / ml), successively at the end of storage period. This means that even after 9 days of cold storage, the counts of

bifidobacteria in all yoghurt treatments were still higher than the minimum level ($10^5 - 10^6$ / ml) that should be present at the consumption of the product to achieve the beneficial effect of bifidobacteria (Hunger and Peitersen, 1992). The decrease of bifidobacterial counts might be due to the development of acidity. Similar trends were obtained by Martin and Choe (1992). Increasing the rate of adding bifidobacteria (*B. bifidum* ATCC 15696, *B. bifidum* ATCC 29521 and *B. longum* BL-04) during probiotic yoghurt manufacture increased the counts of bifidobacteria.

Organoleptic properties:

Scores of probiotic yoghurt organoleptic properties (flavour, body and texture, appearance, acidity and total score) are present in Table (6). Fresh control yoghurt treatments gained the highest scores of organoleptic properties. Also, scores of fresh yoghurt treatments which made by adding bifidobacteria decreased as the amount of bifidobacteria increased. Scores of control treatments decreased slightly as storage period advanced, while scores of treatments made by adding bifidobacteria increased slightly up to sixth day of storage then decreased slightly. Generally, yoghurt treatments made by adding 2.0% of bifidobacteria (T₄, T₇ and T₁₀) gained the highest scores and were not significantly different from control yoghurt treatments. Probiotic yoghurt treatments made by adding *B. bifidum* ATCC 15696 were not significantly different ($p > 0.05$) from corresponding treatments made by adding either *B. bifidum* ATCC 29521 or *B. longum* BL-04, which means that neither the species nor the strain of bifidobacteria affected significantly ($p < 0.05$) the organoleptic properties scores of the resultant probiotic yoghurt. Scores of organoleptic properties of all yoghurt treatments did not significantly ($p > 0.05$) change up to the sixth day of storage period then decreased as storage period progressed. These results are in agreement with those reported by Badawi (2004). Hiroya et al., (1989), found that milk products fermented with mixed starter culture which of *Str. thermophilus* + *L. bulgaricus* + *Bifidobacteria* were given higher scores in the sensory acceptance evaluation than did those fermented with the single starter culture of bifidobacteria.

Part 2: The effect of adding turmeric on the survival of bifidobacteria and organoleptic properties of probiotic yoghurt:

Bifidobacteria count:

Results in Table (7) indicated that the counts of bifidobacteria increased during the first three days of storage period then decreased as storage period advanced in all treatments. Similar results were obtained by **Kebaryet al., (2007)**; **Kebaryet al. (2008)**, the count of bifidobacteria of fresh T₃, T₄, T₅, T₆, T₇ and T₈ were 255, 243, 281, 269, 263 and 271 (cfu × 10⁵ / ml), respectively. Corresponding counts on the sixth day of storage period were 246, 251, 262, 248, 254 and 241 (cfu × 10⁵ / ml), in the same order, but they reached 83, 78, 84, 81, 86 and 81 (cfu × 10⁵ / ml), successively at the end of storage period. This means that even after 12 days of cold storage, the count of bifidobacteria was still higher than the minimum level (10⁵ – 10⁶ / ml) that should be present at the consumption of the product to achieve the beneficial effect of bifidobacteria (**Hunger and Peitersen, 1992**). This decrease might be due to the development of acidity. It could be observed that turmeric concentration did not affect on the survival of bifidobacteria strains. The growth of all bifidobacteria strains was not significantly different from each other.

Organoleptic properties:

Scores of probiotic yoghurt with adding turmeric (1 g / 100 ml milk) organoleptic properties (flavour, body and texture, appearance, acidity and total score) are presented in Table (8). Fresh control yoghurt treatments gained the highest scores of organoleptic properties. Scores of control treatments decreased slightly as storage period advanced, while scores of treatments which made by adding bifidobacteria increased slightly up to the sixth day of storage then decreased slightly. While scores of (T₂, T₄ and T₆) treatments gained lower scores than (T₁) and this due to adding turmeric (1 gm / 100 ml milk). Yoghurt treatments made by adding 2.0% of bifidobacteria (T₃, T₅ and T₇) were not significantly different from control yoghurt treatments (T₁). Scores of organoleptic properties of all yoghurt treatments did not change significantly ($p \leq 0.05$) up to the sixth day of storage period then decreased. These results are in agreement with those reported by **Kebaryet al., (2005)** and **Badawiet al., (2004)**.

Statistical analysis:

Results of statistical analysis of probiotic yoghurt are shown in Table (9&10).

Table (1):Effect of incorporating bifidobacteriaon titratable acidity ofprobiotic yoghurt during storage period

Treatments	Storage period (days)							
	1		3		6		9	
	T.A.%	pH	T.A.%	pH	T.A.%	pH	T.A.%	pH
T ₁ *	0.98	4.75	1.04	4.53	1.09	4.38	1.16	4.26
T ₂	0.87	4.60	0.92	4.56	0.96	4.49	1.06	4.37
T ₃	0.82	4.68	0.87	4.62	0.91	4.55	0.97	4.43
T ₄	0.78	4.71	0.84	4.66	0.89	4.61	0.94	4.46
T ₅	0.90	4.58	0.95	4.55	1.00	4.43	1.09	4.31
T ₆	0.86	4.62	0.93	4.56	0.97	4.49	1.05	4.36
T ₇	0.81	4.66	0.87	4.62	0.91	4.53	0.98	4.41
T ₈	0.87	4.62	0.93	4.57	0.96	4.48	1.06	4.35
T ₉	0.81	4.64	0.89	4.61	0.92	4.52	0.98	4.41
T ₁₀	0.77	4.69	0.85	4.64	0.88	4.58	0.95	4.43

* T₁: yoghurt treatment was made with adding 3.0% normal yoghurt culture.

T₂, T₃ and T₄: yoghurt treatments were made with adding 2, 1.5 and 1.0% normal yoghurt culture and 1.0, 1.5 and 2.0% *Bifidobacteriumbifidum*ATCC 15696, respectively.

T₅, T₆ and T₇: yoghurt treatments were made with adding 2, 1.5 and 1.0% normal yoghurt culture and 1.0, 1.5 and 2.0% *Bifidobacteriumbifidum*ATCC 29521, respectively.

T₈, T₉ and T₁₀: yoghurt treatments were made with adding 2, 1.5 and 1.0% normal yoghurt culture and 1.0, 1.5 and 2.0% *Bifidobacteriumlongum*BL-04, respectively.

Each value in the table is the mean of three replicates.

Table (2):Effect of incorporating bifidobacteria on total solids and diacetyl& acetyl methyl carbinol of probiotic yoghurt during storage period

Treatments*	Storage period (days)							
	1		3		6		9	
	T.S. %	DA + AMC	T.S. %	DA + AMC	T.S. %	DA + AMC	T.S. %	DA + AMC
T₁*	15.69	38.11	15.68	51.06	15.67	68.30	15.72	49.89
T₂	15.66	42.62	15.62	54.21	15.69	62.03	15.68	50.21
T₃	15.71	43.78	15.63	59.22	15.58	69.14	15.66	55.37
T₄	15.67	45.36	15.62	60.21	15.64	74.97	15.67	61.34
T₅	15.68	41.32	15.66	52.12	15.65	66.35	15.67	53.04
T₆	15.66	42.71	15.64	55.72	15.69	70.51	15.66	63.13
T₇	15.69	44.43	15.64	61.34	15.67	76.45	15.70	66.22
T₈	15.69	42.65	15.68	53.78	15.66	68.12	15.71	54.35
T₉	15.67	44.21	15.65	57.32	15.69	72.41	15.68	60.47
T₁₀	15.72	46.71	15.67	62.45	15.67	77.85	15.69	68.32

• See Table (1)

* Each value in the table is the mean of three replicates.

Table (3):Effect of incorporating bifidobacteria on total protein and fat content of probiotic yoghurt during storage period

Treatments	Storage period (days)							
	1		3		6		9	
	Protein	Fat	Protein	Fat	Protein	Fat	Protein	Fat
T₁*	5.56	5.3	5.56	5.5	5.55	5.4	5.55	5.4
T₂	5.54	5.5	5.54	5.5	5.52	5.4	5.50	5.5
T₃	5.52	5.5	5.52	5.6	5.49	5.5	5.48	5.3
T₄	5.55	5.4	5.53	5.4	5.50	5.6	5.52	5.5
T₅	5.53	5.5	5.52	5.5	5.51	5.3	5.50	5.5
T₆	5.57	5.4	5.55	5.5	5.55	5.5	5.54	5.4
T₇	5.55	5.4	5.53	5.3	5.54	5.5	5.52	5.5
T₈	5.52	5.5	5.50	5.5	5.49	5.7	5.49	5.3
T₉	5.51	5.5	5.51	5.4	5.48	5.5	5.48	5.3
T₁₀	5.52	5.3	5.51	5.5	5.51	5.3	5.50	5.5

• See Table (1)

* Each value in the table is the mean of three replicates.

Table (4):Effect of incorporating bifidobacteria on total bacterial and mold & yeast count (cfu / ml x10⁷) of probiotic yoghurt during storage period

Treatments•	Storage period (days)							
	1		3		6		9	
	TBC	Mould & yeast	TBC	Mould & yeast	TBC	Mould & yeast	TBC	Mould & yeast
T₁*	113	ND	127	ND	108	14	87	28
T₂	98	ND	109	ND	82	9	65	16
T₃	82	ND	96	ND	73	6	58	10
T₄	75	ND	83	ND	65	ND	41	6
T₅	102	ND	113	ND	87	6	68	12
T₆	85	ND	96	ND	75	5	56	8
T₇	68	ND	81	ND	66	ND	45	5
T₈	91	ND	102	ND	78	11	63	21
T₉	82	ND	93	ND	75	7	51	9
T₁₀	69	ND	76	ND	61	ND	38	4

• See Table (1)ND: Not detected

* Each value in the table is the mean of three replicates.

Table (5):Effect of incorporating bifidobacteria on the count of bifidobacteria of probiotic yoghurt during storage period

Treatments•	Bifidobacteria counts (CFU / ml) × 10 ⁵			
	Storage period (days)			
	1	3	6	9
T₁*	ND	ND	ND	ND
T₂	173	196	168	113
T₃	205	222	205	152
T₄	223	256	222	176
T₅	187	216	187	126
T₆	213	245	213	168
T₇	242	261	234	191
T₈	156	177	152	109
T₉	192	209	188	148
T₁₀	211	236	209	176

• See Table (1) ND: Not determined

* Each value in the table is the mean of three replicates.

Table (6):Effect of incorporating bifidobacteria on organoleptic properties score of probiotic yoghurt during storage period

Treatments	Organoleptic Properties																			
	Flavour (out of 45)				Body and Texture (out of 35)				Acidity (out of 10)				Appearance (out of 10)				Total (100 Score)			
	1	3	6	9	1	3	6	9	1	3	6	9	1	3	6	9	1	3	6	9
T ₁	42	41	41	40	32	32	30	30	8	8	8	7	9	8	8	7	91	89	87	84
T ₂	40	41	41	39	32	32	32	31	8	8	8	7	8	8	8	8	88	89	89	85
T ₃	40	41	41	40	32	32	31	32	8	8	8	8	8	8	8	7	88	89	88	87
T ₄	40	42	42	42	31	32	32	32	8	8	8	8	7	8	8	7	86	90	90	88
T ₅	42	43	42	40	32	31	32	32	8	8	8	7	8	9	9	8	90	91	91	87
T ₆	42	43	42	40	32	31	31	31	8	8	8	8	8	8	9	8	90	90	90	87
T ₇	41	43	43	42	31	32	32	31	8	8	8	8	7	8	9	8	87	91	92	89
T ₈	40	40	41	39	30	32	32	31	8	8	8	8	7	8	8	7	85	88	89	85
T ₉	40	40	42	40	30	31	32	32	8	8	8	8	7	8	8	7	85	87	90	87
T ₁₀	39	43	42	41	32	32	32	32	7	8	8	8	7	8	8	8	85	91	90	89

● See Table (1).

Each value in the table is the mean of three replicates.

Table(7):Effect of turmeric on viability of bifidobacteria in probiotic yoghurt

Treatments*	Bifidobacteria (CFU × 10 ⁵ / ml)				
	Storage period (days)				
	1	3	6	9	12
T ₁	ND	ND	ND	ND	ND
T ₂	ND	ND	ND	ND	ND
T ₃	255	311	246	171	83
T ₄	243	298	251	163	78
T ₅	281	343	262	183	84
T ₆	269	322	248	178	81
T ₇	263	327	254	167	86
T ₈	271	313	241	173	81

●See Table (1). ND: Not determined

Each value in the table is the mean of three replicates.

Table(8):Effect of turmeric on organoleptic properties score of probiotic yoghurt during storage period

Treatments*	Organoleptic Properties																								
	Flavour (out of 45)					Body and Texture (out of 35)					Acidity (out of 10)					Appearance (out of 10)					Total (100 Score)				
	1	3	6	9	12	1	3	6	9	12	1	3	6	9	12	1	3	6	9	12	1	3	6	9	12
T ₁	42	41	41	38	36	32	32	32	30	30	9	9	8	8	7	9	9	8	7	7	92	91	89	83	80
T ₂	40	39	40	37	35	32	32	32	30	30	9	9	8	8	7	8	8	7	7	6	89	88	87	82	78
T ₃	40	42	41	39	37	30	32	32	31	30	8	9	9	8	8	8	9	9	8	7	86	92	91	86	82
T ₄	39	41	40	38	36	30	32	32	30	29	8	9	9	8	8	8	8	8	7	6	85	90	89	83	79
T ₅	40	42	41	39	37	30	32	32	30	29	8	9	9	8	8	8	9	9	8	7	86	92	91	85	81
T ₆	40	41	39	38	36	30	31	31	30	28	8	9	9	8	8	8	9	9	7	6	86	90	88	83	78
T ₇	40	41	41	39	36	30	32	32	30	29	8	9	9	8	8	8	9	8	7	7	86	91	90	84	80
T ₈	40	41	39	38	36	30	32	32	30	29	8	9	8	8	8	8	9	8	7	6	86	91	87	83	79

• See Table (1).

Each value in the table is the mean of three replicates.

Table (9):Statically analysis of probiotic yoghurt properties

Properties of yoghurt*	Effect of treatments										Effect of storage period (days)					
	Mean squares	Multiple comparisons*										Means squares	Multiple comparisons*			
		T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇	T ₈	T ₉	T ₁₀		1	3	6	9
Titrateable acidity (%)	0.048*	A	C	D	E	B	C	D	C	D	E	0.165*	D	C	B	A
pH value	0.035	E	CD	AB	A	DE	CD	BC	CD	BC	AB	0.387*				
Total solids (%)	2.867	A	A	A	A	A	A	A	A	A	A	9.020	A	A	A	A
Fat (%)	0.014	A	A	A	A	A	A	A	A	A	A	0.021	A	A	A	A
Total protein (%)	7.759	A	A	A	A	A	A	A	A	A	A	0.033	A	A	A	A
Ash (%)	3.575	A	A	A	A	A	A	A	A	A	A	0.017	A	A	A	A
TVFA	74.83*	EF	E	BC	A	D	BC	A	CD	B	A	102.35*	A	B	C	D
DA + AMC (µg/100 ml)	260.23*	E	DE	BC	A	D	B	A	CD	B	A	3502.1*	D	C	A	B
Syneresis (%)	101.542*	A	CD	F	G	B	CD	G	C	EF	G	100.275*	A	C	D	B
Organoleptic properties																
Flavour	8.859*	B	B	AB	A	B	AB	A	B	AB	A	33.204*	A	A	AB	B
Body & Texture	1.573*	AB	B	AB	A	B	AB	A	B	AB	A	8.790*	A	A	AB	B
Appearance	0.401*	A	A	A	A	A	A	A	A	A	A	1.950*	A	A	AB	B
Acidity	1.173*	B	B	AB	A	B	AB	A	B	AB	A	5.341*	A	A	B	C
Total	21.633*	AB	B	AB	A	B	AB	A	B	AB	A	133.500*	A	A	AB	B

• See Table (6).

♦ Each different letter (in the same row) means that multiple comparison are different from each other letter. A is the highest mean followed by B, C, Etc.

* Significant at 0.05

Table (10): Statically analysis of probiotic turmeric yoghurt properties

Properties of yoghurt* Organoleptic properties	Effect of treatments								Effect of storage period (days)						
	Mean squares	Multiple comparisons*								Mean squares	Multiple comparisons*				
		T ₁	T ₂	T ₃	T ₄	T ₅	T ₆	T ₇	T ₈		1	3	6	9	12
Flavour	5.314*	A	B	A	AB	A	AB	A	AB	4.575*	A	A	AB	B	C
Body & Texture	2.571	A	A	A	A	A	A	A	A	22.5*	A	A	AB	B	C
Appearance	0.760*	A	B	A	B	A	B	A	B	16.275*	A	A	AB	B	C
Acidity	2.014	A	A	A	A	A	A	A	A	18.175*	A	A	B	C	C
Total	19.371*	A	B	A	AB	A	AB	A	AB	3.598*	A	A	AB	B	C

• See Table (7).

♦ Each different letter (in the same row) means that multiple comparison are different from each other letter. A is the highest mean followed by B, C, Etc.

* Significant at 0.05.

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تأثير إضافة بكتيريا البافيدوباكتيريوم والكرم على جودة الزبادى

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ملخص البحث :

تم دراسة تأثير إضافة بكتيريا البافيدوباكتيريوم والكرم على جودة الزبادى المنتج بواسطة سلالات مختلفة من بكتيريا البافيدوباكتيريوم والتغيرات الكيميائية والميكروبيولوجية والخواص الحسية أثناء تخزين الزبادى تم دراستها. وأظهرت النتائج أن الزبادى المصنع من البادى الطبيعى كان ذات حموضة أعلى من تلك الأنواع المصنعة من سلالات بكتيريا البافيدوباكتيريوم . كذلك لوحظ حدوث انخفاض فى نسبة الحموضة الكلية فى عينات الزبادى بزيادة نسبة بكتيريا البافيدوباكتيريوم. عينات الزبادى المصنع من سلالات بكتيريا البافيدوباكتيريوم لها قيم الأس الهيدروجيني أعلى من العينة الضابطة . كان هناك ارتباط إيجابي بين نسبة اضافة بكتيريا البافيدوباكتيريوم ودرجة الحموضة . لوحظ عدم تأثير كلا من سلالة و نوع بكتيريا البافيدوباكتيريوم فى محتوى الزبادى من المواد الصلبة الكلية . لوحظ زيادة تدريجية فى محتوى الزبادى من ثنائي الأستيل ميثيل كربينول يزداد تدريجيا حتى اليوم السادس من التخزين ، ثم يحدث انخفاض مع تقدم فترة التخزين . كذلك لوحظ عدم وجود فروق معنوية بين عينات الزبادى فى نسبة كلا من البروتين والدهون . العدد الكلى للبكتيريا فى كل عينات الزبادى يزداد حتى اليوم الثالث من التخزين ثم ينخفض حتى نهاية فترة التخزين . أدى اضافة بكتيريا البافيدوباكتيريوم إلى حدوث انخفاض معنوى فى اعداد الخمائر والفطريات . لوحظ بعد ٩ أيام من التخزين البارد، أن اعداد بكتيريا البافيدوباكتيريوم مازالت مرتفعة فى كل عينات الزبادى ولا تزال أعلى من الحد الأدنى لها (١٠^٦ - ١٠^٧ / مل) ، التي يجب أن تكون موجودة فى الزبادى المنتج لاحداث التأثير المرغوب والفعال . لوحظ ايضا عدم وجود فروق معنوية فى الخواص الحسية لزيادى البروبيوتيك المنتج بواسطة سلالات مختلفة من بكتيريا البافيدوباكتيريوم . كذلك لوحظ بعد ١٢ يوما من التخزين البارد أن اعداد بكتيريا البافيدوباكتيريوم مع الكرم لايزال أعلى من الحد الأدنى للمستوى (١٠^٦ - ١٠^٧ / مل) التي يجب أن تكون موجودة فى المنتج عند الأستهلاك . وجد أن تركيز الكرم ليس له تأثير على حيوية سلالات بكتيريا البافيدوباكتيريوم . وحدث انخفاض بسيط فى الخواص الحسية لعينات الزبادى المحتوية على بكتيريا البافيدوباكتيريوم مع الكرم.

الكلمات الدالة : بكتيريا البافيدوباكتيريوم الكرم جودة الزبادى.