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الاستفادة من الخواص الحيوية لبكتريا البيفيدو المكبسلة والغير مكبسلة فى صناعة الجبن القريش

سامى فاروق محمود

قسم بحوث الألبان – معهد بحوث تكنولوجيا الأغذية – مركز البحوث الزراعية – الجيزة – مصر

الملخص العربى :

يهدف هذا البحث لإضافة سلالات من بكتريا **Bifidobacteria** مثل ***B. bifidum* DI** و ***B. infantis* 4038** إلى الجبن القريش سواء الخلايا الحرة أو تلك المحمية فى كريات من كاراجينات وتأثير هذه المعاملة على حيوية بكتريا **Bifidobacteria** أثناء تخزين الجبن وتأثيرها على خواص وصفات الجبن أثناء تخزينها لمدة ٢٨ يوم . ولقد أوضحت النتائج المتحصل عليها ما يلى :

- لم تُؤثر بكتريا **Bifidobacteria** تأثيراً محسوساً سواء المضافة على صورة خلايا حرة أو تلك المحمية بكريات كاراجينات على كلٍ من الرطوبة أو الدهن أو البروتين الكلى ولكن كان لها تأثير معنوى على كلٍ من الحموضة والـ **pH** والنيتروجين الذائب والخواص الحسية .
- كان تأثير فترة التخزين على الجبن هو انخفاض نسب كلٍ من الرطوبة والبروتين الكلى والـ **pH** والخواص الحسية بينما ازداد كل من الحموضة والنيتروجين الذائب .
- ازداد أعداد بكتريا **Bifidobacteria** حتى اليوم السابع ثم انخفضت أعدادها حيث كان الانخفاض كبيراً فى حالة الجبن المضاف إليها البكتريا فى صورة خلايا حرة حيث انخفضت أعدادها بمقدار ٥ دورات لوغاريتمية فى نهاية فترة التخزين بينما انخفضت أعداد البكتريا فى الجبن المضاف إليها البكتريا المحمية بمقدار ٢ دورة لوغاريتمية .
- احتوت الجبن المضاف إليها بكتريا ***B. bifidum* DI** على أعلى عدد من **Bifidobacteria** يليها بكتريا ***B. infantis* 4038** .
- ازداد العدد الكلى للبكتريا حتى اليوم السابع ثم انخفض تدريجياً حتى نهاية فترة التخزين فى كل المعاملات .
- ازداد أعداد الفطريات والخمائر مع تقدم فترة التخزين زيادة تدريجية وكانت أعلى نتيجة فى نهاية فترة التخزين فى كل المعاملات .

EFFECT OF ADDING CAPSULATED AND UNCAPSULATED BIFIDOBACTERIA ON QUALITY OF KAREISH CHEESE

S. F. Mahmoud

Department of Dairy Res., Food Techn. Res. Institute, Agric. Res. Center, Giza, Egypt.

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ABSTRACT: ***Bifidobacterium bifidum DI and Bifidobacterium infants 4058 either as free cells or entrapped cells in carageenan beads were incorporated separately into Kareish cheese that stored in refrigerator (5 – 7°C) for 28 days. The count of bifidobacteria in cheese made with added free cells decreased pronouncly after the seventh day of storage up to the end of storage period. It decreased by about 5 logarithmic cycle within 21 days, while the entrapped cells decreased by only about 2 logarithmic cycles during the same period. Entrapment of bifidobacteria cells in alginate beads improved their survival during storage of Kareish cheese. Cheese made with adding B. bifidum DI contained the highest count of bifidobacteria followed by B. infantis 4038. Total bacterial count increased up to the seventh day then decreased gradually in all Kareish cheese up to the end of storage period. Mould & yeast count increased in all cheese treatments during storage period. Incorporation of bifidobacteria into Kareish cheese did not affect significantly the chemical composition except acidity, pH value and soluble nitrogen content. There were significant differences among cheese treatments in scores of organoleptic properties in all cheese treatments. Moisture content, total nitrogen content, pH value and scores of Kareish cheese were decrease as storage period progressed, while acidity and soluble nitrogen contents increased.***

Key words: ***Kareish cheese, bifidobacteria, free cells, entrapped cells, carageenan beads.***

INTRODUCTION

Kareish cheese is one of soft cheeses which are most popular in Egypt and Arabian countries owing to its high protein, low fat and reasonable price. It is an acid coagulated fresh cheese, made from skim milk with soft composition, white curd and slightly salty. Kareish cheese is considered one of the most food production rich in calcium and phosphorus. These elements are essential for bones and teeth formation. It is also rich in sodium and potassium, which play an important role in the formation of body liquids and muscles (Francois *et al.,* 2004).

Bifidobacteria are the predominant gut flora in breastfed infant (Rasic and Kurmann, 1983). These bacteria are becoming recognized worldwide because of

their health and nutritional benefits such as potential beneficial roles in human intestinal tract (Kurmann and Rasic, 1991; Robinson and Samona, 1992). Antitumorigenic activity, improvement of lactose tolerance, reduction of serum cholesterol levels, reduction of ammonia and free serum phenol in patients with liver disease, synthesis of vitamins, increased immunocompetence and antagonistic effects towards enteropathogenic bacteria have all been deviled (Anand *et al.,* 1984; Kageyama *et al.,* 1984; Yamazak *et al.,* 1985; Kebary, 1995 and Badawi and El-Sonbaty, 1997). It is estimated that over 70 products containing bifidobacteria are produced worldwide (Shah, 1997). They include fermented milk, butter milk, sour cream, frozen dessert, cheese, baby foods, pharmaceutical preparations and livestock feed supplements (Gomes *et al.,* 1995; Tamime *et al.,* 1995; Kebary, 1996; Kebary *et al.,* 1998; Badawi and Hussein 1999). Microencapsulation has been used to improve the viability of bifidobacteria in dairy products (Dinakar and Mistry, 1994; Kebary *et al.,* 1998; Hussein and Kebary, 1998).

In view of the aforementioned the objectives of this study were to investigate the possibility of incorporating microentrapped bifidobacteria in Kareish cheese, study the effect of microentrapment on viability of bifidobacteria, study the behavior of different strains of bifidobacteria during cold storage of Kareish cheese and to investigate the effect of bifidobacteria on cheese quality.

MATERIALS AND METHODS

Materials:

Buffalo's skim milk was obtained from the Faculty of Agriculture, Cairo University, Egypt. Pure culture of *Str. thermophilus* and *Lb. bulgaricus* was obtained from Hansen Laboratories (Denmark). *Bifidobacterium bifidum* DI was provided by Diversitech Inc. (Gainesville, FL), while *Bifidobacterium infantis* 4038 was provided by Prof. Khamis Kebary (Dairy Sci. Department, Faculty of Agriculture, Minufiya University, Egypt). Sodium chloride was obtained from the local market.

Methods:

1. Preparation of microencapsulated bifidobacteria:

Microencapsulation of bifidobacterial cells were prepared by the method of Adhikari *et al.* (2000) as following: Bifidobacterial cells were grown in MRS broth containing L-cystein- HCl, maintained at 37°C for 24 h. the cells were harvested by centrifugation at 5000 rpm, washed twice in sterile normal saline under the same centrifugation conditions and resuspended in 10 ml of sterile normal saline. A 2% K-carageenan solution containing 0.9% NaCl (to improve dispersability of the K-carageenan) was prepared and heat treated at 96°C for 6 min. Sixty ml of K-carageenan solution was thoroughly mixed with

20 ml of cell suspension, and temporarily kept in a water bath at 47 $\pm$ °C. Ten ml of soybean oil containing 0.1% Tween 80 was tempered by stirring to 40°C on a stirrer hotplate for 2 to 3 min. the mixture of cells and K-carageenan was then quickly added with continuous stirring to the oil in the beaker and the resultant mixture was further stirred for about 10 min. to allow for emulsification and encapsulation to occur. The emulsion was removed by the addition of 150 ml of sterile 0.3 M KCL. After that, the oil phase was removed from the top of the mixture with a sterilized separator funnel under the laminar flow, the capsules were harvested from the KCL solution by gentle centrifugation at 350 xg for 10 min, the capsules were washed twice with 0.3 M KCL for better stability under the same centrifugation condition and finally stored in refrigerator before use.

2. Cheese making:

Buffalo's skim milk was heated to 85°C. Milk was divided into 5 batches, one of them was served as control (2% normal starter), to another two batches un capsulated cells of *Bifidobacterium bifidum* DI and *Bifidobacterium infantis* 4038 were added separately at the rate $\simeq$ 1.0 × 10^7 cfu/ ml milk plus 1.0% normal starter. The Kareish cheese was manufactured as described by Effat *et al.* (2001). The cheeses were packed in plastic bags contained salted whey and stored in refrigerator at 5 – 7°C for 4 weeks and analyzed at 0, 7, 14, 21 and 28 days.

Chemical analysis:

Fresh Kareish cheese samples analyzed for titratable acidity (TA) and soluble nitrogen (SN) as described by Ling (1963). The pH value was measured using pH meter (HANNA 8417). Moisture, fat and total protein were determined as described by A.O.A.C (2000).

Microbiological analysis:

All samples were examined for total bacterial counts (TBC) and moulds and yeasts according to the American Public Health Association (APHA, 1992). Bifidobacteria were enumerated on modified MRS agar (Ventling and Mistry, 1993) with NNL solution (neomycin sulphate 0.2%, nalidixic acid 0.03% and Lithium chloride 6.0%) (Samona and Robinson, 1991). Samples containing beads were suspended in 9.0 ml sterile phosphate buffer (1 m, pH 7.5) followed by gentle shaking at room temperature for 10 min to release bifidobacteria from beads (Sheu *et al.,* 1993).

Sensory evaluation:

The sensory evaluation of Kareish cheeses was carried out according to El-Shafei *et al.* (2008). The samples were presented to the panelists in a

random order. The cheeses were evaluated organoleptically after zero, 14 and 28 days of storage period in Dairy Science Department, Food Technology Research Institute, Agriculture Research Center. Panelists evaluated cheese for appearance (20 points), body and texture (35 points), flavor (45 points) and overall acceptability (100 points).

Statistical analysis:

2 × 2 factorial design was used to analyze the data and Duncan's test was used to make the multiple comparisons (Steel and Torrie, 1980). Significant differences were determined at $p \leq 0.05$ level.

RESULTS AND DISCUSSION

Moisture content of all cheese batches decreased ($p \leq 0.05$) gradually during the first 14 days of storage, then decreased slightly up to the end of storage period (Table 1). This might be due to the development of acidity, which leads to curd contraction that helps to expel the whey from the curd (Effat *et al.,* 2001). Adding bifidobacteria during manufacturing of Kareish cheese either as free cells or entrapped cells did not affect significantly ($p > 0.05$) the moisture content of the resultant cheese (Table 1). Also the species of bifidobacteria did not affect significant ($p > 0.05$) the moisture content of Kareish cheese (Table 1).

Fat content of all Kareish cheese treatments decreased slightly during storage period (Table 1) (Mohamed *et al.,* 2009). There were no significant ($p > 0.05$) differences among cheese treatments, which means neither the species nor the entrapment affected significantly ($p > 0.05$) the fat content of the resultant cheese. Total protein content of Kareish cheese decreased significantly ($p \leq 0.05$) as storage period progressed (Table 1). This decrease may be due to the loss of water soluble nitrogen compounds results from the protein degradation during storage in whey (El-Shafei *et al.,* 2008). Adding bifidobacteria during manufacturing of Kareish cheese did not have significant ($p > 0.05$) effect on total protein content of Kareish cheese (Table 1).

Titratable acidity of all cheese treatments increased significantly ($p \leq 0.05$) as storage period progressed (Table 2). This increase in acidity could be due to conversion of the residual lactose in cheese by starter bacteria. Similar results were reported by Badawi and Kebary (1998) and Farag *et al.* (1988). Incorporation of bifidobacteria especially the free cells caused a significant ($p \leq 0.05$) increase in acidity of Kareish cheese. Cheese contained free cells of bifidobacteria had higher acidity than those contained entrapped bifidobacteria (Table 2) which might be due to the availability of lactose in the medium (Badawi and Hussein, 1999).

Table 1

Table 2

pH values of cheese treatments as affected by adding bifidobacteria, the form of adding these bacteria and the storage period followed an opposite trend to those of titratable acidity (Table 2).

Soluble nitrogen (SN) content increased significantly ($p \leq 0.05$) as storage period progressed. SN increased Markedly during the first 14 days then increased gradually up to the end of storage period. These results are in agreement with those reported for Tallaga and Domiati cheese by Farag *et al.* (1988); Kebary (1995) and Badawi and Kebary (1996 & 1998). Cheese treatments contained free cells of bifidobacteria had higher soluble nitrogen than those contained entrapped bifidobacteria (Table 2).

The behavior of bifidobacteria during refrigerated storage of Kareish cheese are presented in Table (3). Bifidobacteria were not detected in the control. Counts of bifidobacteria in all cheeses treatments made with adding either free cells (unentrapped) or entrapped cells increased up to the seventh day of refrigerated storage with different growth rates. The results agree with those of Gomes and Malcata (1998) and Badawi and Hussein (1999). Cheese treatments made with adding free cells exhibited higher counts of bifidobacteria than those made with entrapped cells in the beginning, which might be due to the availability of the nutrients. The survival of bifidobacteria in all cheeses made with adding bifidobacteria decreased after the seventh day up to the end of storage period (Blanchette *et al.,* 1996). The viability of bifidobacteria in cheeses made with added free cells bifidobacteria decreased markedly from 38×10^7, 40×10^7 at the seventh day to 60×10^2, 51×10^2 cfu/g at the end of storage period for *B. bifidum* DI and *B. infants* 4038, respectively (Table 3). These results revealed that the viable counts of bifidobacteria in cheeses made with adding free cells decreased by about 5 logarithmic cycles during 21 days which might be due to the effect of salt and refrigerated temperature on bifidobacteria. On the other hand, entrapment of bifidobacteria in carageenan beads improved pronouncedly the survival of bifidobacteria during refrigerated storage of Kareish cheese (Table 3). The counts of bifidobacteria in cheeses made with added entrapped bifidobacteria cells (*B. bifidum* DI and *B. infants* 4038) decreased by about 2 logarithmic cycles. These results indicated that entrapment of bifidobacteria in carageenan beads protected the cells from the effect of salt and cold storage.

Total bacterial counts of all Kareish cheese increased up to the seventh day of refrigerated storage, then decreased gradually up to the end of cold storage (Table 4). This decrease might be due to the low temperature, developing acidity and the effect of salt. Similar results were reported by Mohamed *et al.* (2009). Cheeses that contained higher counts of bifidobacteria contained lower total bacterial counts (Table 4). The results

Table 3

Table 4

show that the mould and yeast increased gradually during storage period and reached its maximum by the end of cold storage period for all Kareish cheese treatments. Generally the control and cheeses made with free cells of bifidobacteria contained higher mould and yeast than those made with entrapment bifidobacteria scores given for flavor, body & texture and appearance of Kareish cheese are presented in Table (5). All cheeses were accepted by panelists even at the end of storage period.

The scores of flavor, body & texture and appearance decreased slightly after 14 days of refrigerated storage. Cheese treatments made by adding encapsulated bifidobacteria gained higher scores than those of the corresponding cheese treatments made by adding free cells of bifidobacteria. It could be concluded that adding bifidobacteria did not affect the chemical composition of Kareish, while increased the acceptability of cheese. Encapsulation of bifidobacteria improved their survival. Cheese treatment made by adding entrapped bifidobacteria even after cold storage for 28 days contained higher counts of bifidobacteria than should be present to achieve the therapeutic effect.

Table 5

REFERENCES

Adhikari, K., A. Mustapha and I. U. Grun (2000). Viability of Micro-encapsulated bifidobacteria in set yoghurt during refrigerated storage. J. Dairy Sci., 83: 1946 – 1951.

Anand, S. K., R. A. Srinrasan and L. K. Rao (1984). Antibacterial activity associated with *Bifidobacterium longum*. Cult. Dairy Prod. J., 19: 6 – 8.

A.O.A.C. (2000). Association of Official Analytical Chemists. Official Methods of Analysis, 25th Ed., Arlington, VA, USA.

APHA, American Public Health Association, Standard Methods for the Examination of Dairy Products 1992, Amer. Publ. Health Assoc. Inc. 12th Ed., New York.

Badawi, R. M. and A. H. El-Sonbaty (1997). Viability of *Staphylococcus aureus* and *Escherichia coli* in zabady made with bifidobacteria. Egyptian J. Dairy Sci., 25 (2): 217 – 233.

Badawi, R. M. and S. A. Hussein (1999). Survival of microentrapped Bifidobacteria during storage of white cheese and their effect on cheese quality. Minufiya J. Agric. Res., 24 (2): 493 – 513.

Badawi, R. M. and K. M. K. Kebary (1996). Accelerated ripening of cheese by partial lyzed lactococci. Minufiya J. Agric. Res., 21 (1): 63-81.

Badawi, R. M. and K. M. K. Kebary (1998). Influence of fat replacers on the quality of low fat Tallaga cheese. Proc. 7th Egyptian Conf. Dairy Sci. and Techn., pp. 347-365.

Blanchette, L., D. Roy, G. Belanger and S. F. Gauthier (1996). Production of cottage cheese using dressing fermented by bifidobacteria. J. Dairy Sci., 79: 8 – 15.

Dinakar, P. and V. V. Mistry (1994). Growth and viability of *Bifidobacterium bifidum* in Cheddar cheese. J. Dairy Sci., 77: 2854 – 2864.

Effat, B. A., M. M. E. Salem and K. El-Shafei (2001). Effect of using different starters on quality of Kareish cheese. Egypt J. Food Sci., 29: 95 – 108.

El-Shafei, Kawther, M.; A. M. Abd El-Gawad, N. Dabiza, O. M. Shraf and B. A. Effat (2008). A mixed culture of *Propionibacterium thoenii* p-127, *Lactobacillus rhamnosus* and *Lactobacillus plantrum* as protective culture in Kareish cheese. Pol. J. Food Nutr. Sci., 58 (4): 433 – 441.

Farag, Seham I., Nafisa A. El-Saify and A. I. Hamed (1988). Microbiological and chemical studies on pickled Domiati cheese treated with some ripening factors. Minufiya J. Agric. Res., 13: 1845.

Francois, Z. N., N. Ahmed, M. T. Felicite and M. El-Soda (2004). Effect of ropy and capsular exopolysaccha rides producing strain of *Lactobacillus plantarum* 162 RM on characteristics and functionality of fermented milk and soft Kareish type cheese. African J. Biotechnol., 3: 512 – 518.

Gomes, A. M. P. and F. X. Malcata (1998). Development of probiotic cheese manufactured from goat milk: response surface analysis via technological

manipulation. J. Dairy Sci., 81: 1492 – 1507.

Gomes, Ana M. P., F. X. Malcata, F. A. M. Klaver and H. Grande (1995). Incorporation and survival of *Bifidobacterium* sp. strain Bo and *Lactobacillus acidophilus* strain Ki in a cheese product. Neth. Milk Dairy J., 49: 71 – 95.

Hussein, S. A. and K. M. K. Kebary (1998). Improving viability of bifidobacteria by microentrapment and their effect on some pathogenic bacteria in stirred yoghurt. Acta Alimentaria. Accepted.

Kageyama, T., T. Tomoda and Y. Nakano (1984). The effect of bifidobacteria administration in patient with leukemia. Bif. Microflora, 3 (1): 29 – 36.

Kebary, K. M. K. (1995). Production, partial purification and stability of antimicrobial substances produced by *Bifidobacterium bifidum* DI. Egyptian J. Dairy Sci., 23: 151 – 166.

Kebary, K. M. K. (1996). Viability of *Bifidobacterium bifidum* and its effect on quality of frozen Zabady. Food Res. Int., 29: 431 – 437.

Kebary, K. M. K., S. A. Hussein and R. M. Badawi (1998). Improving viability of bifidobacteria in frozen ice milk. Egyptian J. Dairy Sci., 26: 319 – 337.

Kurmann, J. A. and J. L. Rasic (1991). The health potential of products containing bifidobacteria. In Therapeutic Properties of Fermented Milk, ed. R. K. Robinson. Elsevier Applied Science, London.

Ling, E. R. (1963). A text book of dairy chemistry, vol. II, Chapan and Hall, Ltd., London.

Mohamed, Ragia O., Mona E. Youssef and Amany A. Salem (2009). Effect of some Medicinal Herbs on the quality and shelflife of Kareish cheese. Egyptian J. Agric., 87 (5): 1463 – 1477.

Râsic, J. L. and J. A. Kurmann (1983). Bifidobacteria and their roles. Birkhauser Berlag., Basel, Switzerland.

Robinson, R. K. and A. Samona (1992). Health aspects of Bifidus products: A review. Int. J. Food Sci. Nutrition, 43: 175 – 180.

Samona, A. and R. K. Robinson (1991). Enumeration of bifidobacteria in dairy products. J. Soc. Dairy Technol., 44: 64 – 66.

Shah, N. P. (1997). Bifidobacteria: Characteristics and potential for application in fermented milk products. Milchwissen-schaft, 52 (1): 16 – 21.

Sheu, T. Y., R. T. Marshal and H. Heyman (1993). Improving survival of culture bacteria in frozen desserts by microentrapment. J. Dairy Sci., 76 (6): 1902 – 1907.

Steel, R. G. D. and J. H. Torrie (1980). Principles and Procedures of Statistics; A Biometrical Approach, 2nd Edn. McGraw-Hill, New York.

Tamime, A. Y., V. M. E. Marshall and R. K. Robinson (1995). Microbiological and technological aspects of mikls fermented by bifidobacteria. J. Dairy Res., 62: 151 – 187.

Ventling, B. L. and V. V. Mistry (1993). Growth characteristics of bifidobacteria in ultrafiltered milk. J. Dairy Sci., 76: 962 – 971.

Yamazaki, S., K. Machii, S. Tsuyuki, H. Momose, T. Kawashima and K. Ogasa (1985). Immunological responses to monoassociated *Bifidobacterium longum* and their relation to prevention of bacterial invasion. Immunol., 56: 43 – 51.

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سامى فاروق محمود

قسم بحوث الألبان – معهد بحوث تكنولوجيا الأغذية – مركز البحوث الزراعية – الجيزة – مصر

الملخص العربى :

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- لم تُؤثر بكتريا **Bifidobacteria** تأثيراً محسوساً سواء المضافة على صورة خلايا حرة أو تلك المحمية بكريات كاراجينات على كلٍ من الرطوبة أو الدهن أو البروتين الكلى ولكن كان لها تأثير معنوى على كلٍ من الحموضة والـ **pH** والنيتروجين الذائب والخواص الحسية .
- كان تأثير فترة التخزين على الجبن هو انخفاض نسب كلٍ من الرطوبة والبروتين الكلى والـ **pH** والخواص الحسية بينما ازداد كل من الحموضة والنيتروجين الذائب .
- ازداد أعداد بكتريا **Bifidobacteria** حتى اليوم السابع ثم انخفضت أعدادها حيث كان الانخفاض كبيراً فى حالة الجبن المضاف إليها البكتريا فى صورة خلايا حرة حيث انخفضت أعدادها بمقدار ٥ دورات لوغاريتمية فى نهاية فترة التخزين بينما انخفضت أعداد البكتريا فى الجبن المضاف إليها البكتريا المحمية بمقدار ٢ دورة لوغاريتمية .
- احتوت الجبن المضاف إليها بكتريا ***B. bifidum*** **DI** على أعلى عدد من **Bifidobacteria** يليها بكتريا ***B. infantis*** **4038** .
- ازداد العدد الكلى للبكتريا حتى اليوم السابع ثم انخفض تدريجياً حتى نهاية فترة التخزين فى كل المعاملات .
- ازداد أعداد الفطريات والخمائر مع تقدم فترة التخزين زيادة تدريجية وكانت أعلى نتيجة فى نهاية فترة التخزين فى كل المعاملات .

Table (1). Moisture, fat and total protein contents of Kareish cheese made with adding bifidobacteria during refrigerated storage.

Cheese treatments	Moisture content (%)					Fat content (%)					Total protein content (%)				
	Storage period					Storage period					Storage period				
	0	7	14	21	28	0	7	14	21	28	0	7	14	21	28
C	76.55^{Aa}	75.20^{Ab}	74.12^{Ac}	73.66^{Ad}	73.00^{Ad}	0.60^{Aa}	0.60^{Aa}	0.60^{Aa}	0.50^{Aa}	0.50^{Aa}	16.50^{Aa}	16.02^{Ab}	15.50^{Ac}	15.45^{Ac}	15.40^{Ac}
D	76.50^{Aa}	75.22^{Ab}	74.10^{Ac}	73.60^{Acd}	72.88^{Ad}	0.60^{Aa}	0.60^{Aa}	0.60^{Aa}	0.60^{Aa}	0.60^{Aa}	16.52^{Aa}	16.00^{Ab}	15.55^{Ac}	15.46^{Ac}	15.41^{Ac}
DA	76.60^{Aa}	75.30^{Ab}	74.20^{Ac}	73.58^{Ac}	72.88^{Acd}	0.60^{Aa}	0.60^{Aa}	0.60^{Aa}	0.60^{Aa}	0.50^{Aa}	16.48^{Aa}	15.97^{Ab}	15.52^{Ac}	15.42^{Ac}	15.38^{Ac}
I	76.66^{Aa}	75.28^{Ab}	74.28^{Ac}	73.62^{Ac}	73.10^{Acd}	0.60^{Aa}	0.60^{Aa}	0.60^{Aa}	0.60^{Aa}	0.50^{Aa}	16.55^{Aa}	16.08^{Ab}	15.58^{Ac}	15.49^{Ac}	15.45^{Ac}
IA	76.62^{Aa}	75.25^{Ab}	74.24^{Ac}	73.55^{Acd}	73.08^{Ad}	0.60^{Aa}	0.60^{Aa}	0.60^{Aa}	0.60^{Aa}	0.60^{Aa}	16.54^{Aa}	16.07^{Ab}	15.59^{Ac}	15.50^{Ac}	15.43^{Ac}

Capital letters for the differences in the same column, while small letters for the differences in the same row. Letter A is the highest mean followed by B, C, … etc. significant at 0.05 level.
C: Cheese treatment made by normal starter (control).
D: Cheese treatment made by normal starter plus *Bifidobacterium bifidum* DI (free cells).
DA: Cheese treatment made by normal starter plus *Bifidobacterium bifidum* DI (encapsulated).
I: Cheese treatment made by normal starter plus *Bifidobacterium infantis* (free cells).
IA: Cheese treatment made by normal starter plus *Bifidobacterium infantis* (encapsulated).

Table (2). Titratable acidity, pH values and soluble nitrogen contents of Kareish cheese made with adding bifidobacteria during refrigerated storage.

Cheese treatments*	Titratable acidity content (%)					pH value					Soluble nitrogen content (%)				
	Storage period					Storage period					Storage period				
	0	7	14	21	28	0	7	14	21	28	0	7	14	21	28
C	0.95^{Ae}	1.10^{Ad}	1.20^{Ac}	1.28^{Ab}	1.31^{Aa}	5.30^{Ba}	5.20^{Cb}	5.08^{Cc}	5.00^{Cd}	4.98^{Be}	0.40^{Ac}	0.58^{Ad}	0.81^{Ac}	0.90^{Ab}	1.03^{Aa}
D	0.90^{Be}	1.06^{Bd}	1.15^{Bc}	1.25^{Bb}	1.30^{Aa}	5.32^{Ba}	5.23^{Bb}	5.11^{Bc}	5.03^{Bd}	5.00^{Be}	0.41^{Ae}	0.57^{ABd}	0.78^{ABc}	0.91^{Ab}	1.02^{Aa}
DA	0.85^{Ce}	1.01^{Cd}	1.08^{Cc}	1.10^{Cb}	1.25^{Ba}	5.40^{Aa}	5.31^{Ab}	5.17^{Ac}	5.11^{Ad}	5.09^{Ae}	0.35^{Be}	0.51^{Bd}	0.69^{Bc}	0.79^{Bb}	0.93^{Ba}
I	0.92^{Be}	1.07^{Bd}	1.17^{Bc}	1.26^{Bb}	1.30^{Aa}	5.32^{Ba}	5.24^{Bb}	5.12^{Bc}	5.05^{Bd}	5.01^{Be}	0.42^{Ae}	0.59^{Ad}	0.80^{Ac}	0.92^{Ab}	1.05^{Aa}
IA	0.86^{Ce}	1.00^{Cd}	1.07^{Cc}	1.11^{Cb}	1.24^{Ba}	5.41^{Aa}	5.31^{Ab}	5.18^{Ac}	5.10^{Ad}	5.08^{Ae}	0.32^{Ce}	0.47^{Cd}	0.67^{BCc}	0.76^{Cb}	0.92^{Ba}

Capital letters for the differences in the same column, while small letters for the differences in the same row. Letter A is the highest mean followed by B, C, … etc. significant at 0.05 level.
*** See Table (1).**

Table (3). Effect of microentrapment on survival of bifidobacteria during refrigerated storage of Kareish cheese made with adding bifidobacteria during refrigerated storage.

Cheese treatments*	Viable counts of bifidobacteria (cfu / g)				
	Storage period				
	0	7	14	21	28
C	ND	ND	ND	ND	ND
D	20×10^{6}	38×10^{7}	50×10^{4}	70×10^{3}	60×10^{2}
DA	10×10^{6}	20×10^{7}	15×10^{6}	80×10^{5}	40×10^{5}
I	22×10^{6}	40×10^{7}	46×10^{4}	60×10^{3}	51×10^{2}
IA	15×10^{6}	22×10^{7}	17×10^{6}	75×10^{5}	32×10^{5}

ND: not detected.
* See Table (1).

Table (4). Total bacterial counts and mould and yeast of Kareish cheese made with adding bifidobacteria during refrigerated storage.

Cheese treatments*	Total bacterial counts × 10^5					Mould and yeast × 10^2				
	Storage period					Storage period				
	0	7	14	21	28	0	7	14	21	28
C	490	540	96	90	75	12	14	24	30	35
D	450	510	90	80	62	13	20	31	42	50
DA	400	460	60	50	35	12	16	25	32	37
I	452	515	85	70	60	15	26	38	52	60
IA	390	455	62	48	30	12	15	26	33	38

* See Table (1).

Table (5). Sensory evaluation of Kareish cheese made with adding bifidobacteria during refrigerated storage.

Cheese treatments*	Flavour (45)			Body and texture (35)			Appearance (20)			Total score (100)		
	Storage period (days)			Storage period (days)			Storage period (days)			Storage period (days)		
	0	14	28	0	14	28	0	14	28	0	14	28
C	40^{Aa}	38^{ABab}	35^{Bbc}	31^{Aa}	30^{Aa}	27^{Bb}	19^{Aa}	18^{Aab}	17^{Ab}	90^{Aa}	86^{Bb}	79^{Cc}
D	40^{Aa}	36^{Bb}	34^{Bc}	31^{Aa}	30^{Aa}	26^{Bb}	19^{Aa}	19^{Aa}	17^{Ab}	90^{Aa}	85^{Bb}	77^{Dc}
DA	40^{Aa}	40^{Aa}	38^{Ab}	31^{Aa}	30^{Aa}	30^{Aa}	19^{Aa}	19^{Aa}	18^{Aa}	90^{Aa}	89^{Aa}	86^{Ab}
I	39^{Aa}	37^{Bb}	35^{Bc}	30^{Aa}	29^{Aa}	29^{Aa}	19^{Aa}	18^{Aab}	17^{Ab}	88^{Aa}	84^{Bb}	81^{Cc}
IA	40^{Aa}	38^{ABb}	37^{Ab}	31^{Aa}	30^{Aa}	30^{Aa}	19^{Aa}	18^{Aab}	17^{Ab}	90^{Aa}	86^{Bb}	84^{Bc}

Capital letters for the differences in the same column, while small letters for the differences in the same row. Letter A is the highest mean followed by B, C, … etc. significant at 0.05 level.

*** See Table (1).**