VIABILITY AND METABOLIC ACTIVITY OF MICROENCAPSULATED BIFIDOBACTERIA IN PLAIN AND STRAWBERRY STIRRED YOGHURT

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Abstract

Viable count of microencapsulated *Bifidobacterium bifidum Bifidobacterium lactis* and *Bifidobacterium longum* were mixed with plain and strawberry stirred yoghurt and refrigerated storage for two weeks. Subsequent viability of *Bifidobacterium* species, level of fermentation end products (lactic and acetic acids) and pH values were measured during storage period. The results showed that the population of encapsulated bifidobacteria were more stable (percentage of viable count 53 – 80%) than that, in nonencapsulated (percentage of viable count 2 – 14%), at the end of storage period. The encapsulated *Bifidobacterium lactis* in plain yoghurt recorded 83 and 80% of viable count after first and second week from storage period. On the other hand, survivals of all encapsulated *Bifidobacterium* species were higher in plain yoghurt than that in strawberry yoghurt. It is worthy to notice that, the encapsulated bifidobacteria release a little metabolic end products than nonencapsulated. Results suggested that microencapsulation protected bifidobacteria from the low pH of yoghurt and strawberry yoghurt.

INTRODUCTION

Bifidobacteria, one of the most important probiotic bacteria, Schrezenmeir and DeVrese (2001) recently defined the word " probiotic" as a preparation of or a product containing viable, defined microorganisms in sufficient number, which alters the microflora (by implantation or colonization) in a compartment of the host and by that, exert beneficial health in this host. These probiotic organisms can potentially provide beneficial effects for fermented products consumers. Examples of these potential benefits include improving lactose digestion (Kim and Gilliland 1983), control of serum cholesterol levels (Gilliland *et al.* 1985, De Rodas *et al.* 1996, Anderson and Gilliland 1999), stated anti-cancer activities, and stimulation of immune system (Schrezenmeir and DeVrese, 2001). The ultimate intent of this approach is to provide the gastrointestinal tract of humans with elevated viable populations of bifidobacteria (Coussement 1996). It is worthy to note that Kurmann

et al. (1992), mentioned that, 10^6- 10^7 probiotic organisms per day would ensure transit of viable bacteria through the stomach. Thus, viability of bifidobacteria much attention. However, the survival of bifidobacteria in yoghurt is quite low because of the low pH of this product (4.2 to 4.6) and the presence of lactic and acetic acids, which are known as antimicrobial agents (Lankaputhra *et al.* 1996). So, the effect of low pH, addition of sugar and/or acidic fruits and cold storage on viability of bifidobacteri have been studied by many investigators. On the other hand, Reuter (1990) recorded a significant decrease colony-forming unit of 3 to 4 log cycles of bifidobacteria during storage in yoghurt-like product. Accordingly, Saleh (1997), observed as mush as a 1 log cycle (90%) reduction of numbers of viable bifidobacteria in strawberry set soy-yoghurt after 12 day of refrigerated storage. Moreover, Lankaputhra *et al.* (1996) stated that viability of bifidobacteria strains such as *Bifidobacterium infantis* in 12 % skim milk at pH 4.3 was decreased by 30 % after 12 day of storage at 4°C. Besides, after 24 day at the same temperature, the counts decreased by more than 82%. Medina and Jordano (1994) found a 93% reduction in bifidobacterial counts of fermented milk produced in Spain at 7°C. So that using the mechanism of microencapsulation to enhance the viability of bifidobacteria in plain and strawberry stirred yoghurt. Therefore, this study was undertaken to apply bifidobacteria in plain and strawberry stirred yoghurt as encapsulated cells. Our objective were to determined the survival and metabolic activity (production of lactic and acetic acid) of encapsulated and free of three bifidobacteria strains under high stress of plain and strawberry stirred yoghurt environments (low pH, acidic fruits, sugars and cold storage) during storage.

MATERIALS AND METHODS

1. Materials:

1.1. Milk: Skim buffalo milk, were obtained from Food Technology Research Institute, Agricultural Research Center

1.2. Cultures: Lyophilized yoghurt culture for direct vat set "DVS" type *Lactobacillius dulbrueekii* sub sp. *bulgaricus* and *Streptococcus salivarius* sub sp. *thermophilus* (YC-180), as well as a freeze-dried culture for direct vat set "DVS" type *Bifidobacterium bifidum* Bb-11, *Bifidobacterium lactis* Bb-12, and *Bifidobacterium longum* Bb-46 were supplied by Chr. Hansen Laboratories, Copenhagen, Denmark.

2. Methods:

2.1. Preparation of microencapsulated bifidobacteria: Microencapsulation of bifidobacterial cells were prepared by the method of Adhikari *et al.* (2003) 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% κ-carageenan solution containing 0.9% NaCl (to improve dispersability of the κ-carageenan) was prepared and heat treated at 96°C for 6 min. Sixty mL of κ-carageenan solution was thoroughly mixed with 20 ml of cell suspension, and temporarily kept in a water bath at 47±°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 κ - 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.

2.2. Preparation of strawberry juice: Strawberry juice was produced as mentioned by Arbuckle (1977) from fresh fruit as follows: fruit was washed and mixed with sugar in the ratio of 3:1 (w/w) fruit to cane sugar and held at about 5°C for 12 hours. Then the mixture was homogenized using tempest homogenizer at 5000 rpm and stored in the deep freezer until used.

2.3. Manufacture of yoghurt treatments: Yoghurt was made according to the method of (Tamime and Robinson 1985). Skim buffalo milk was heated at 85°C for 10 min and cooled to 43°C. Milk was inculcated with approximately 0.07% (w/v) of lyophilized yoghurt culture (YC-180). The mixture was then, incubated at 42°C until

the final pH decreased to 4.5. Yoghurt was stirred and divided into 14 portions, as follows:

1) Plain stirred yoghurt without bifidobacteria (control)
2) Strawberry stirred yoghurt without bifidobacteria (control)
3) Plain stirred yoghurt containing 0.07% (w/v) free *B. bifidum* cells (lyophilized)
4) Plain stirred yoghurt containing 0.07% (w/v) free *B. lactis* cells (lyophilized)
5) Plain stirred yoghurt containing 0.07% (w/v) free *B. longum* cells (lyophilized)
6) Strawberry stirred yoghurt containing 0.07% (w/v) free *B. bifidum* cells (lyophilized)
7) Strawberry stirred yoghurt containing 0.07% (w/v) free *B. lactis* cells (lyophilized)
8) Strawberry stirred yoghurt containing 0.07% (w/v) free *B. longum* cells (lyophilized)
9) Plain stirred yoghurt containing 10% (w/v) microencapsulated *B. bifidum*
10) Plain stirred yoghurt containing 10% (w/v) microencapsulated *B. lactis*
11) Plain stirred yoghurt containing 10% (w/v) microencapsulated *B. longum*
12) Strawberry stirred yoghurt containing 10% (w/v) microencapsulated *B. bifidum*
13) Strawberry stirred yoghurt containing 10% (w/v) microencapsulated *B. lactis*
14) Strawberry stirred yoghurt containing 10% (w/v) microencapsulated *B. longum*

The above treatments mentioned were stored in refrigerator for two weeks.

2.4. Bifidobacterial count: Ten grams of yoghurt treatments containing microencapsulated bifidobacteria were mixed with 90 mL of 0.05 M EDTA solution made in 0.1 M sodium phosphate buffer (pH 7), to release the cells from the capsules and incubating at 42°C for 20 min according to Adhikari *et al.* (2003). The count of released bifidobacteria and in the other treatments (yoghurt containing free bifidobacteria) was determined according to Dinakar & Mistry (1994). The mixture of antibiotics including 2 g of paromomycin sulfate, 0.3 g of nalidixic acid, and 60 g of lithium chloride, was prepared in 1L of distilled water, filter-sterilized 0.2µm, and stored at 4°C until use. The mixture of antibiotics (5ml) was added to 100 ml of MRS-Agar medium to inhibit the other bacteria except bifidobacteria. L.cysteine-HCl

(Sigma chemical Co., St. Louis, Mo) was added at the rate of 0.5%, to decrease the redox potential of the medium. Plates were incubated anaerobically at 37°C for 48h.

2.5. Determination of organic acids by HPLC: Organic acids in yoghurt treatments were determined by HPLC according to the method of Adhikari et al., (2000) as follows: Ten grams of sample was mixed with 10 ml of 0.01N H2SO4 and centrifuged at 10000 rpm for 25min and the supernatant was filtrated through a 0.2 µm Millipore membrane filter then 1-3 ml was collected in a vial for injection into HPLC Hewllet Packared (series 1050) equipped with autosampling injector, solvent degasser, ultra violet (UV) detector set at 210 nm and quarter HP pump (series 1050). The column temperature was maintained at 35°C. An isocratic separation was carried out with 0.01N H2SO4 as a mobile phase at flow rate of 1 ml/min. The organic acids standard (lactic and acetic acid) from Fluka Co. were dissolved in a mobile phase and injected into HPLC. Retention time and peak area were used to calculation of organic acid concentration by the data analysis of Hewllet Packared software.

2.6. Sensory evaluation: Sensory evaluation of plain and strawberry stirred yoghurt containing free and microencapsulated *Bifidobacterium lactis* (for example) were carried out by a regular score panel according to Tamime and Robinson (1985).

2.7. Statistical analysis: Data are presented as means and standard deviation. The significant differences among yoghurt treatments were evaluated using a one-way ANOVA to analyze the points of sensory evaluation by Least Significant Different (LSD) at $P < 0.05$

RESULTS AND DISCUSSION

1- Changes in pH values of plain and strawberry stirred yoghurt containing free or microencapsulated bifidobacteria during refrigerated storage period:

The effect of plain and strawberry stirred yoghurt containing free or microencapsulated *Bifidobacterium bifidum*, *Bifidobacterium lactis* and

bifidobacterium longum on the changes of pH values during refrigerated storage period for two weeks is shown in Table (1). The addition of strawberry decreased the pH values when compared with plain yoghurt during the refrigerated storage period. In the all treatments, the pH values were decreased during storage period. The pH values of plain stirred yoghurt containing free or in microencapsulated bifidobacteria strains (zero time) were slightly varied (from 4.42 to 4.45). However, the decrease in pH were almost identical for yoghurt (control) and yoghurt containing microencapsulated bifidobacteria. The pH values were decreased in the case of nonencapsulated treatments of tested strains when compared with capsulated bacteria at the end of storage period. The greater decrease in pH observed in the treatments containing free bifidobacterial cells, this may be due to the double metabolic activities of bifidobacteria and yoghurt culture and the stopping of metabolic activity of capsulated bifidobacteria.

2- Changes in lactic and acetic acids of plain and strawberry stirred yoghurt containing free or microencapsulated bifidobacteria during refrigerated storage period:

From Table (2), the data shows that, the amount of lactic and acetic acids for all tested cultures were increased by progressing the storage period. Production of lactic acid was higher in plain than in strawberry stirred yoghurt in three investigated bifidobacteria strains (free or encapsulated) at the end of refrigerated storage period, this my be due to the antibacterial effect of strawberry (low pH) and sugar content. Sneath (1986) found that the optimum pH for initial growth raged from 6.5 to 7.0 no growth happend at pH 4.5 or 8.0. In plain stirred yoghurt, the maximum amount of lactic acid was recorded by *Bifidobacteium lactis*, while *Bifidobacterium bifidum* produce the highest amount of acetic acid after 2 weeks during the refrigerated storage. However, changes in acetic and lactic acids of microencapsulated cells were similar to those observed with control either in plain or in strawberry yoghurt. On the other hand, the plain yoghurt containing tested microencapsulated bifidobacteria, produce small amount of acetic acid compared

with that containing free cells. These results reveled a little metabolic activity by microencapsulated bifidobacteria. These findings in accordance to that found by Adhikari *et al.* (2000),they reported that the acetic acid content in the yoghurt with nonencapsulated bifidobacteria was higher than that in the plain yoghurt and encapsulated treatments.

3- Viability of microcapsulated bifidobacteria in plain and strawberry stirred yoghurt during refrigerated storage period:

The effect of microcapsulation on the viability of bifidobacteria in plain and strawberry stirred yoghurt during refrigerated storage period was showed in Table (3). The populations of bifidobacteria free cells in plain and strawberry yoghurt were decline sharply during refrigerated storage period. Moreover, strawberry yoghurt recorded a minimum of viable count of bifidobacteria compared with that in plain yoghurt either containing free or microencapsulated bifidobacteria. On the other hand, the population of encapsulated *Bifidobacterium* strains were more stable (percentage of viable count 53 – 80%) than that in nonencapsulated (percentage of viable count 2 – 14%), at the end of storage period. The encapsulated *Bifidobacterium lactis* in plain yoghurt recorded 82.63 and 80% of viable count after first and second week, respectively from storage period. While the viable percentage of encapsulated *Bifidobacterium longum* were 83.63 and 61.46% at the same period. Whereas, survival of all encapsulated *Bifidobacterium* species were higher in plain yoghurt than that in flavored yoghurt. These results were in agreement with Khalil and Mansour (1998) they found that the viability of bifidobacteria encapsulated was increased in calcium alginate were incorporated in mayonnaise, and storage studies were done for 16 week. Also, Dinaker and Mistry (1994), reported that the microencapsulaion has been shown to be effective method for maintaining the viability of probiotic cultures in yoghurt. On the other hand, Lee and Heo (2000), found that the death rate of *Bifidobacterium longum* in the calcium alginate beads decreased proportionally with an increase in both the alginate gel concentration and bead size. Adhikari *et al.* (2003), reported that the

decline in population of *Bifidobactrium longum,* irrespective of strain, was significant from 5th day, there was a reduction of 89.3% (0.97 log cycles) and 91.8% (1.08 log cycles)in the population of *B. longum* B6 and *B. longum* ATCC 15708, respectively, in yoghurt containing nonencapsulated cells over the storage period. In a parallel line, Godward and Kailasapathy (2003) recorded that encapsulation followed by freeze-drying of the probiotic bacterial calls ensured the smallest reduction in the viability of cells in yoghurt. From the pervious results, we can observe that the microencapsulation protected the bifidobacteria in plain and strawberry yoghurt during refrigerated storage.

4- Sensory evaluation:

Table (4) showed the sensory evaluation of plain and strawberry stirred yoghurt containing free and microencapsulated bifidobacteria. The results of statistical analysis explained significant difference between the taste of plain yoghurt containing free and microencapsulated bifidobacteria when compared with plain yoghurt (plain control), but the microencapsulated bifidobacteria in plain yoghurt which gave score (42.5) is good, this is in agreement with results reported by (Tamime and Robinson 1985). On the other hand, no significant differences between the taste of strawberry yoghurt (strawberry control) and other treatments. Whereas, the other characteristics (appearance, consistency and smell) score exhibited no significant difference between the plain and strawberry yoghurt containing free and microencapsulated bifidobacteria and their controls. From the statistical analysis of sensory evaluation, that preparation of yoghurt containing microencapsulated bifidobacteria are accepted.

Table 1. Changes in pH values of plain and strawberry stirred yoghurt containing free or microencapsulated bifidobacteria during refrigerated storage period.

Storage period (week)	Control□		Free cells						Microencapsulated cells					
			B. bifidum		*B. lactis*		*B. longum*		*B. bifidum*		*B. lactis*		*B. longum*	
	Plain‡	Straw*	Plain	Straw	Plain	Straw	Plain	Straw	Plain	Straw	Plain	Straw	Plain	Straw
	← pH values →													
0	4.44	4.30	4.44	4.30	4.43	4.30	4.47	4.31	4.43	4.29	4.42	4.37	4.45	4.28
1	4.38	4.26	4.32	4.25	4.30	4.25	4.33	4.26	4.39	4.27	4.37	4.26	4.36	4.27
2	4.33	4.15	4.22	4.16	4.22	4.14	4.28	4.22	4.32	4.17	4.28	4.14	4.28	4.22

□ Stirred yoghurt with out bifidobacteria

‡ Plain stirred yoghurt

* Strawberry stirred yoghurt

Table 2. Changes in lactic and acetic acids of plain and strawberry stirred yoghurt containing free or microencapsulated bifidobacteria during refrigerated storage period.

Strains	Storage period (week)	Control□				Free cells				Microencapsulated cells			
		Plain‡		Strawberry*		Plain		Strawberry		Plain		Strawberry	
		Organic acids (mg/100g)											
		Lactic	Acetic	Lactic	Acetic	Lactic	Acetic	Lactic	Acetic	Lactic	Acetic	Lactic	Acetic
B. bifidum	0	599.9	29.5	450.3	55.3	622.7	35.9	459.5	78.5	606.9	29.7	468.3	63.5
	1	682.3	35.9	511.1	72.8	652.5	55.0	510.5	128.7	634.2	42.4	506.4	107.9
	2	697.6	45.3	563.0	81.4	741.5	147.5	560.1	61.5	672.1	44.6	541.2	58.4
B. lactis	0					635.6	35.7	503.5	95.5	636.7	21.9	501.4	97.4
	1					714.9	86.6	611.2	101.3	903.8	27.4	625.4	100.5
	2					801.7	102.5	655.3	28.3	631.3	34.2	577.7	17.6
B. longum	0					641.4	57.9	551.1	25.3	599.0	33.5	545.0	13.4
	1					677.9	59.7	621.5	53.4	610.6	43.4	601.5	50.3
	2					655.6	61.2	653.5	87.9	728.8	48.5	715.0	56.6

□ Stirred yoghurt with out bifidobacteria

‡ Plain stirred yoghurt

* Strawberry stirred yoghurt

Table 3. Effect of microencapsulation on the viability of bifidobacteria in plain and strawberry stirred yoghurt during refrigerated storage period.

Strains	Storage period (week)	Free cells				Microencapsulated cells			
		Plain‡		Strawberry*		Plain		Strawberry	
		Count□	Viability %ψ	Count	Viability %	Count	Viability %	Count	Viability %
B. bifidum	0	203	100	203	100	381	100	340	100
	1	19	9.36	13	6.40	288	75.60	207	60.88
	2	9	4.43	4	1.79	254	66.67	180	52.94
B. lactis	0	216	100	260	100	380	100	267	100
	1	40	18.52	21	8.08	314	82.63	200	74.91
	2	20	9.26	11	4.23	304	80.00	150	56.18
B. longum	0	250	100	233	100	397	100	270	100
	1	45	18.00	17	7.30	332	83.63	202	74.81
	2	35	14.00	10	4.29	244	61.46	150	55.55

‡ Plain stirred yoghurt

* Strawberry stirred yoghurt

□ $\log_7$ CFU/g of stirred yoghurt

ψ Viability % = (CFU after storage period /initial CFU) × 100

Table 4. Sensory evaluation of plain and strawberry stirred yoghurt containing free and microencapsulated bifidobacteria

Characteristics	Maximum points	Plain‡			Strawberry Ψ		
		Control□	Free cells	Microencapsulated cells	Control□	Free cells	Microencapsulated cells
Taste	50	48.0±1.2	41.5±6.9*	42.5±4.8*	47.0±1.6	43.9±5.1	43.8±9.7
Appearance	20	18.3±0.8	16.9±2.6	17.6±3.2	18.7±0.9	18.0±2.2	18.9±2.6
Consistency	20	16.3±0.7	16.9±3.0	17.1±2.3	17.3±1.2	16.2±2.3	19.1±9.4
Small or odor	10	8.7±0.9	8.7±1.7	8.3±1.6	8.9±1.0	8.6±1.1	8.3±1.9

* The mean is significant at $P< 0.05$

□ Stirred yoghurt with out bifidobacteria

‡ Plain stirred yoghurt

Ψ Strawberry stirred yoghurt

REFERENCES

1. Adhikari, K., Mustapha, A. and Grun, I. U. 2000. Viability of micro-encapsulated bifidobacteria in set yogurt during refrigerated storage. J. Dairy Sci 83: 1946-1951.

2. Anderson, J. W. and Gilliland, S. E. 1999. Effects of fermented milk (yoghurt) containing *Lactobacillus acidophilus* L1 on serum cholesterol in hypercholesterolemic humans. J. Am. Coll. Nutr. 18 (1): 43-50.

3. Arbuklc, W. S. 1977. Ice cream. 3rd Edition pub. By the A VI Publishing Co., INC, West port. Connecticut, USA.

4. Coussement, P. 1996. Pre-and synbiotics with inulin and oligofructose: promising developments in functional foods. Food Tech Eur. (Dec-Jan): 102-104.

5. De Rodas, B. Z., Gilliland, S. E. and Maxwell, C. V. 1996. Hypo-cholesterolemic action of *Lactobacillus acidophilus* ATTC 43121 and cacium in swine with hypercholesterolemia anduced by diet. J. Dairy Sci. 79 (12): 2121-2128.

6. Dinaker, P. and Mistry, V. V. 1994. Growth and viability of *Bifidobacterium bifidum* in Chedder cheese. J Dairy Sci. 77 (10): 2854-2864.

7. Gilliland, S. E. Nelson, C. R. and Maxwell, C. 1985. Assimilation of cholesterol by *Lactobacillus acidophilus.* Appl. Environ. Microbiol. 49 (2): 377-381.

8. Godward, G. and Kailasapathy, K. 2003. Viability and survival of free encapsulated and co-encapsulated probiotic bacteria in yoghurt. Milchwissenschaft 58 (7/8) 396-399.

9. Khalil, A. H. and Mansour, E. H. 1998. Alginate encapsulated bifidobacteria survival in mayonnaise. J. Food Sci. 63 (4): 702-705.

10. Kim, H. S. and Gilliland, S. E. 1983. *Lactobacillus acidophilus* as adietary for milk to aid lactose digestion in humans. J. Dairy Sci. 66 (5): 959-966.

11. Kurmann. J. A., Rasic J. L. and Kroger, M. 1992. Encyclopedia of fermented fresh milk product. Pub. Van Nostrand, Reinhold, New York.

12. Lankaputhra, W. E. V., Shah, N. and Britz, M. 1996. Survival of bifidobacteria during refrigerated storage in presence of acid and hydrogen peroxide. Michwiss 17 (2): 65-70.

13. Lee, K. Y. and Heo, T. R. 2000. Survival of *Bifidobacterium longum* immobilized in calcium alginate beads in simulated gastric juices and bile salt solution. Applied and environmental Microbiology, February, (66): 869-873.

14. Medina, L. M. and Jordano, R. 1994. Survival of constitutive microflora in commercially fermented milk containing bifidobacteria during refrigerated storage. J. Food Protection 56:731-733.

15. Reuter, G. 1990. Bifidobacteria culture as components of yoghurt-like products. Bifidobacteria Microflora 9, 107-113

16. Saleh, F. A. 1997. Studies on the bacterial fermentation of soymilk and its effect on the production of soy milk-based yoghurt. MSc Dairy Sci. and Tech. Dep. Faculty of Agri. Cairo Univ.

17. Schreenmeir, J. and De Vrese, M. 2001. Probiotics, prebioticas and symbiotics to approaching a definition. Am. J. Clin. Nutr. 73 (Supp): 361s-364s.

18. Sneath, P. H. A, Mair, N. S, Elisabeth Sharpe, M. and Holt, J. G. 1986. Bergeys manual of systematic bacteriology. Vol. 2. Williams & Wilkins, London.

19. Tamime, A. Y. and Robinson, R. K. 1985. Yoghurt, Science and Tech- nology. Paramon press, Oxford, England.

القدرة على الحياة والنشاط الميتابوليزمى لبكتيريا البيفيدو المكبسلة الموجودة فى الزبادي المقلب العادي والزبادي بالفراولة

فرج على صالح – سحر مصطفى كامل – نبيه عبد الحميد إبراهيم

قسم الأغذية الخاصة والتغذية-معهد بحوث تكنولوجيا الأغذية-مركز البحوث الزراعية

تم خلط عدد حى من ثلاث انواع من بكتيريا البيفيدو *Bifidobacterium bifidum* و*Bifidobacterium lactis* و *Bifidobacterium longum* المكبسلة (وضع بكتيريا البيفيدو داخل كبسولة دقيقة) مع الزبادى العادى والزبادى بالفراولة المقلب ثم التخزين فى الثلاجة لمدة أسبوعين لقياس قدرة هذه البكتيريا على الحياة ومستوى نشاطها الميتابوليزمى الناتج من عملية التخمير (حامض اللاكتيك وحامض الاستيك) وأيضا قياس رقم الـ pH أثناء التخزين. و أظهرت النتائج أن عدد بكتيريا البيفيدو المكبسلة كانت أكثر ثباتا (نسبة العدد الحى يتراوح بين ٥٣-٨٠%) بالمقارنة بالبكتيريا الحرة والتى كان نسبة العدد الحى فيها يتراوح بين ٢-١٤% و ذلك فى نهاية مدة التخزين. وسجلت بكتيريا البيفيدو وبكتيريا *B.lactis* نسبة بقاء حى ٨٣ و ٨٠% بعد الأسبوع الأول والثاني على التوالي من التخزين . ومن جهة أخرى كان العدد الحي لكل أنواع بكتيريا البيفيدو المكبسلة والمختبرة أعلى فى الزبادي العادي عن الزبادى بالفراولة.ومن الجدير بالذكر أن بكتيريا البيفيدو المكبسلة كانت لها نسبة نشاط ميتابوليزمى قليل عن البكتيريا الحرة. ومن النتائج يتبين أن عملية الكبسلة تقى هذه البكتيريا من الظروف المحيطة والتى تتمثل فى انخفاض رقم ال pH فى كل من الزبادى العادى والزبادى بالفراولة.