

Research Article

Encapsulation of Different Types of Probiotic Bacteria within Conventional/Multilayer Emulsion and Its Effect on the Properties of Probiotic Yogurt

Hamed Mahmoodi Pour,¹ Mohammad Hossein Marhamatizadeh ,¹ and Hossein Fattahi²

¹Department of Food Hygiene, Faculty of Veterinary Medicine, Kazeroon Branch, Islamic Azad University, Kazeroon, Iran

²Department of Microbiology, Faculty of Veterinary Medicine, Kazeroon Branch, Islamic Azad University, Kazeroon, Iran

Correspondence should be addressed to Mohammad Hossein Marhamatizadeh; drmarhamati@gmail.com

Received 15 April 2021; Revised 26 February 2022; Accepted 15 March 2022; Published 31 March 2022

Academic Editor: Antimo Di Maro

Copyright © 2022 Hamed Mahmoodi Pour et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Microencapsulation of probiotic cells within emulsion is an efficient method to enhance the viability of probiotic bacteria. In the present study, free and encapsulated probiotic cells (*Lactobacillus rhamnosus* and *Lactobacillus plantarum*) in simple and multilayer emulsions were used to produce a set of probiotic yogurts. In all samples, an increasing trend in syneresis and acidity values and a decreasing trend in pH and viability of probiotic cells were observed during the storage time. However, the changes in these parameters were more significant for free-loaded probiotic samples. Moreover, the free cells showed poor survival in the yogurt samples by decreasing the viable cell count of probiotics from 7.71–7.59 logs CFU/mL to 6.93–6.82 log CFU/mL during storage, while encapsulation in the multilayer emulsion showed an insignificant reduction from 7.65–7.59 logs CFU/mL to 7.55–7.45 log CFU/mL at the end of storage. The obtained results showed that the type of probiotic bacteria had no significant effects on the physicochemical and structural properties of samples. However, encapsulating probiotics in multilayer emulsion led to a more homogenous structure in yogurt. The sensorial properties were also not affected by the probiotic type and the encapsulation method. Consequently, the multilayer emulsion can provide an ideal delivery carrier for encapsulating probiotic bacteria in dairy products.

1. Introduction

Functional food refers to food with positive effects on the human body by lowering the risk of diseases. Recently, the demands for foods containing functional elements (e.g., probiotics) with positive impacts on preventing, controlling, or healing different health problems have increased. The new lifestyle and using ready-to-eat industrial foods and fast foods have increased the risk of disease. In this regard, probiotic-loaded foods can be considered functional foods that affect consumer health by lowering blood cholesterol and fat, developing body immunity and mineral absorption, preventing and controlling cancer, and treating gastric ulcer [1–3]. Therefore, people are looking for healthy and functional foods such as probiotics and/or synbiotic foods [4, 5].

The presence of live probiotic microorganisms (MOs) at high enough concentrations (more than 10^7 CFU/mL) in food products causes beneficial health effects on consumers [6, 7]. These types of food products, which are known as probiotic foods, affect the consumer's health by lowering blood cholesterol and fat, developing body immunity and mineral absorption, preventing and controlling cancer, and treating gastric ulcer. The predominant bacteria in probiotic foods is *Lactobacillus* [8, 9]. The viability and concentration of the probiotic MOs in the food products before consumption and at the expiry date is an important parameter. The type of starter culture, the strain of probiotic MO, the condition of storage, and the concentration of lactic acid and oxygen are the main parameters that can influence the viability of MOs in probiotic yogurts [10]. Therefore, keeping

the concentration and viability of probiotic MOs up to a certain level has become an important challenge in developing probiotic yogurts [11, 12]. One of the main strategies for enhancing the MOs viability is using prebiotic carbohydrates in the food formulation. These types of foods which contain probiotic MOs and prebiotic agents are known as synbiotics [8]. Encapsulation of probiotics is also investigated recently to extend their viability and functionality during storage. To this end, Afzaal et al. [13] studied the viability of encapsulated probiotic bacteria in yogurt. Muzzafar and Sharma [14] evaluated the microencapsulation of probiotics for incorporation in cream biscuits. Chen et al. [15] evaluated the effect of xanthan-chitosan-xanthan double layer encapsulation on survival of *Bifidobacterium* BB01 in yogurt.

The main objective of the present work was evaluating the impacts of probiotic type (*Lactobacillus rhamnosus* and *Lactobacillus plantarum*) and encapsulation method using simple and multilayer emulsions on the cell viability and physicochemical, rheological, structural, and sensorial properties of yogurts.

2. Materials and Methods

2.1. Materials. Tween 80 and MRS agar were bought from Merck Company (Darmstadt, Germany). Whey protein isolate (WPI, 98% whey protein) was purchased from BiPRO; Davisco Foods Intl., Eden Prairie, MN. Persian gum was purchased from Dena Emulsion Company (Shiraz, Iran). Olive oil was purchased from a local market in Shiraz.

2.2. Preparation of Bacteria. Viable *L. rhamnosus* and *L. plantarum* cultures were prepared as described in our previous work [10]. Briefly, 0.5 mL MRS broth was poured into each tube containing the freeze-dried cultures. The

obtained suspension was added to 20 mL MRS broth and then maintained at 37°C for 18h. After that, the vial was centrifuged at 4000 rpm for 180 s, and the obtained sediment was collected. All these actions were performed in a sterile environment.

2.3. Emulsion Preparation

2.3.1. Convective Emulsion. To prepare conventional emulsion, Tween 80 (2.5%w/w) was added to distilled water (DW, 87.5%w/w). The olive oil (9%w/w) containing a pellet of bacteria (*L. rhamnosus* or *L. plantarum*, 1% w/w) was added to the aqueous phase and mixed well at 15000 rpm for 240 s by a high-speed homogenizer (Ultra-Turrax T18, IKA, Germany).

2.3.2. Multilayer Emulsion. WPI (2 g) was added to DW (57.5 g) and hydrated for 12 h. Olive oil (9 g w/w) containing a pellet of bacteria (*L. rhamnosus* or *L. plantarum*, 1% w/w) was added to the prepared aqueous phase and mixed at 15000 rpm for 120 s by a high-speed homogenizer (Ultra-Turrax T18, IKA, Germany). Then, 30 g of Persian gum dispersion (3.3%, w/w) was added to the prepared emulsion and homogenized at the same condition to obtain multilayer emulsion.

2.3.3. Encapsulation Efficiency. To determine the encapsulation efficiency (EE) of the probiotic bacteria in the prepared emulsions, 10 g of each sample was centrifuged at 260 g (20 min at 4°C), followed by separating the water phase. The bacterial count in the sample and the separated water phase was then determined, and EE was calculated using the following equation:

$$EE (\%) = \frac{(\text{Bacterial count of emulsion} - \text{Bacterial count of water phase})}{\text{Bacterial count of emulsion}} \quad (1)$$

2.4. Yogurt Preparation. Stirred yogurt samples were produced based on the method explained by Fazilah et al. [16] with some modifications. First, milk was heated at 85°C for half an hour and then cooled down to 42°C. The starter culture (YoFlexO express 1.0, Chr. Hansen Inc., Milwaukee, WI) including *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp *bulgaricus* was added to milk and inoculated at 42°C. Six different samples were prepared by adding six different probiotic bacteria (free or encapsulated) to the milk: 1. LR (free *L. rhamnosus*), 2. LR-ME (*L. rhamnosus* encapsulated within multilayer emulsion), 3. LR-E (*L. rhamnosus* encapsulated within conventional emulsion), 4. LP (free *L. plantarum*), 5. LP-ME (*L. plantarum* encapsulated within multilayer emulsion), and 6. LP-E (*L. plantarum* encapsulated within emulsion). The prepared mixtures were poured into the cups and incubated at 42 ± 1°C until reaching pH 4.2 ± 0.1. The yogurt samples were cooled and kept at 4 ± 1°C for

21 days. The percentage of dry matter (12.5%) and fat (2.5%) were relatively constant in all samples.

2.4.1. Fat Determination. The fat content of the yogurts was determined based on the AACC standard method [17].

2.4.2. pH and Acidity. Total titratable acidity and pH of yogurt samples were determined based on AOAC standard method [18]. Titratable acidity was reported as the percentage of lactic acid every 7 days over 21 days of the refrigeration storage period.

2.4.3. Determination of Probiotic Bacteria Viability. To specify the number of viable probiotic bacteria in the yogurts, the number of *L. rhamnosus* and *L. plantarum*

colonies were counted on MRS agar after inoculating MRS agar with each of the samples and incubating at $37 \pm 1^\circ\text{C}$ for 3 days under an anaerobic condition [19]. The number of lactic acid bacteria was reported during 21 days of the refrigeration storage period. In addition, the starter culture count was determined based on the previous standard method [20].

2.4.4. Viscosity Measurement. The viscosity of the yogurt samples was measured by a Brookfield cone and plate viscometer (DV2 Pro II, Brookfield Engineering Laboratories, USA) at 21°C . The applied shear rate was 80.64 s^{-1} , and the spindle type was CP51 [21].

2.4.5. Syneresis Measurement. Syneresis measurements were carried out according to the method of Gilbert et al. [22] with some modifications. Each sample was weighed, equilibrated at 10°C for 120 min, and centrifuged at 10°C ($238 \times g$, 10 min). The expelled serum was then collected and weighed. The ratio between the expelled serum and the yogurt mass was reported as syneresis.

2.4.6. Color Measurement. Brightness (L^*), redness-greenness (a^*), and yellowness-blueness (b^*) of the surface of samples were determined using a digital camera (Canon PowerShot A540, 6 megapixels resolution) as reported by Ścibisz et al. [23]. Each sample was placed in a box ($50 \times 50 \times 60 \text{ cm}^3$) equipped with a natural daylight source (6500 K), and the camera was placed on the top of the box at the distance of 25 cm from the sample. L^* , a^* , and b^* were calculated by Adobe Photoshop® CS6.

2.4.7. Surface Electron Micrograph. The surface of each lyophilized yogurt was studied by a scanning electron microscope (TESCAN Vega3, Czech Republic). Micrographs of samples were taken at an accelerating voltage of 20.0 kV after coating samples with a gold layer by a sputter coater (Desk Sputter Coater DSRI, Nanostructural Coating Co., Iran).

2.4.8. Sensory Properties. Sensory evaluation was conducted by 10 well-trained panelists from Pegah Fars Company (Shiraz, Iran). This test was performed according to the 5-point standard hedonic method (1 = dislike extremely, 2 = dislike moderately, 3 = neither like nor dislike, 4 = like moderately, and 5 = like extremely). For this purpose, the samples were kept at 4°C for 1 week prior to sensory tests. The prepared yogurts with random codes were given to the panelists in a white plastic cup to evaluate the characteristics including aroma, taste, color, texture, strength, and general acceptance [24]. Sensory evaluation was conducted in testing booths with different light sources: day light illumination for color assessment and red light for other organoleptic properties. Panelists rinsed their mouths with tap water before tasting and also between each sample.

2.5. Statistical Analysis. All tests were done in the form of completely random blocks and repeated 3 times. Comparison of means was done with Duncan's multiple range test at the significance level of 5%. Data were reported as mean \pm standard deviation. Additionally, the quantitative data were statistically analyzed using SAS Statistical Software, version 9.1 (SAS Institute Inc., 2000; Cary, NC, USA).

3. Results and Discussion

3.1. Encapsulation Efficiency of Emulsions. An effective encapsulation system for probiotics should improve the viability of probiotics and protect them from harsh conditions during processing and gastrointestinal digestion. The encapsulation efficiency of LR-ME, LR-E, LP-ME, and LP-E samples were 98.43 ± 0.42 , 99.02 ± 0.31 , 97.89 ± 0.89 , and $98.05 \pm 0.66\%$, respectively. The results showed that there were no significant differences between the viability of probiotic bacteria in different samples.

3.2. Fat Content. The amount of fat in yogurt can affect the pH value, acidity, viscosity, structure, and sensorial properties [20]. The results showed that the probiotic and encapsulation type had no significant effects on the fat content of yogurt samples (Figure 1). The fat contents of samples were in the range of 2.51 to 2.54%. Similar results were also reported by Ningtyas et al. [25] and Ranadheera et al. [26].

3.3. pH. Yogurt production has relied on the activity of two homofermentative bacteria as well as the production of lactic acid and other aromatic compounds. The production of acid is the main reason for coagulation during fermentation. One of the main reasons for the unilateral selection of starters for yogurt preparation is their ability for acid generation during 4 to 5 h of incubation [20]. The survival of probiotic bacteria in foods is influenced by the pH of the food. The pH of yogurt during storage is shown in Figure 2. Based on the results, the pH values of all samples significantly ($p < 0.05$) reduced during storage. Similarly, Tseng and Zhao [27] reported a pH reduction during the storage of yogurts. The results attained by Temiz et al. [28] and Turgut and Cakmakci [29] also found that the pH of fruit yogurts decreases during storage. According to Figure 2, the samples containing free bacteria showed lower pH values than those with encapsulated bacteria. In addition, among probiotic bacteria, *L. rhamnosus* had stronger effects on pH reduction. The effect of free probiotic bacteria on reducing postacidification was more pronounced compared to the encapsulated ones [14, 29]. The pH reduction might be attributed to the production of β -galactosidase enzyme by the starters. Moreover, the application of remaining carbohydrates by survived microorganisms over the storage time can lead to the production of lactic acid, formic acid, and CO_2 [30].

3.4. Acidity. The titratable acidity values of functional yogurt samples are shown in Figure 3. The acidity of all functional yogurts has been significantly increased with a stable pace

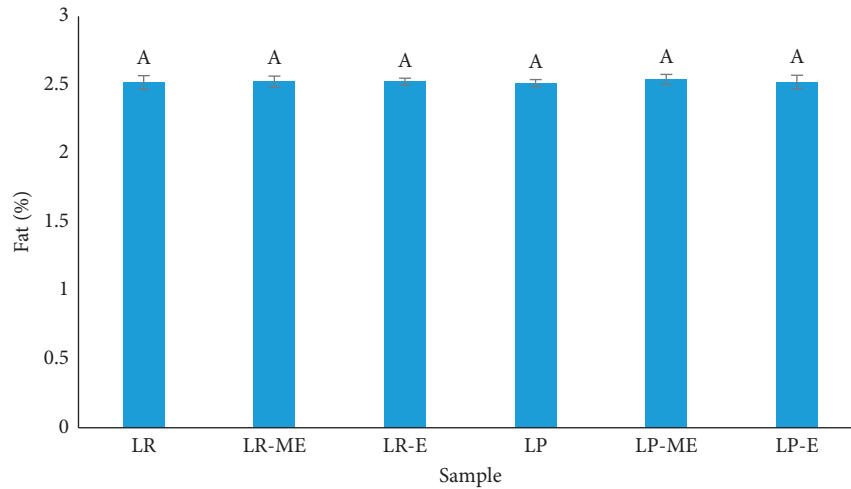


FIGURE 1: Fat content of probiotic yogurt. LR (*L. rhamnosus*), LR-ME (encapsulated *L. rhamnosus* in multilayer emulsion), LR-E (encapsulated *L. rhamnosus* in emulsion), LP (*L. plantarum*), LP-ME (encapsulated *L. plantarum* in multilayer emulsion), and LP-E (encapsulated *L. plantarum* in emulsion).

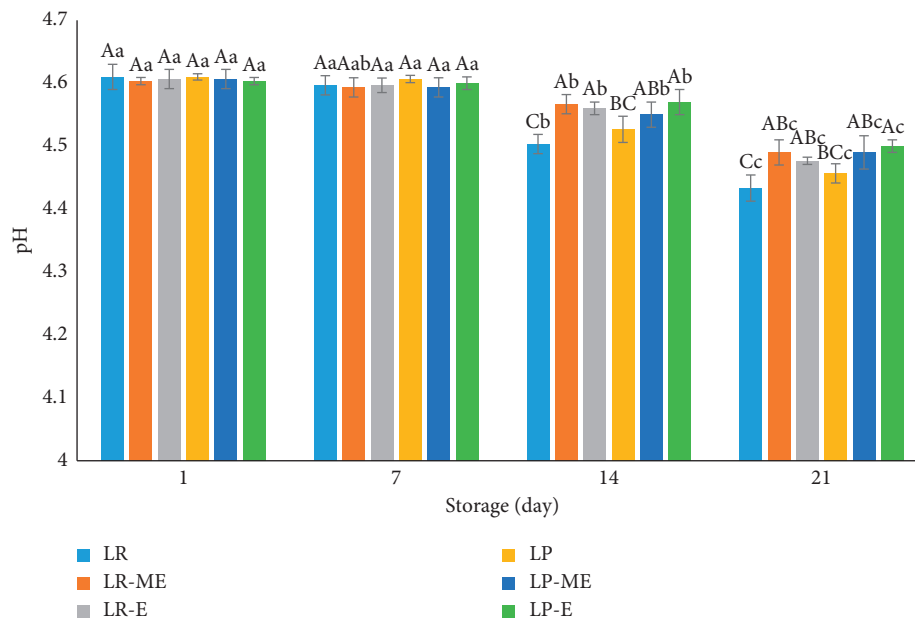


FIGURE 2: pH content of probiotic yogurt. LR (*L. rhamnosus*), LR-ME (encapsulated *L. rhamnosus* in multilayer emulsion), LR-E (encapsulated *L. rhamnosus* in emulsion), LP (*L. plantarum*), LP-ME (encapsulated *L. plantarum* in multilayer emulsion), and LP-E (encapsulated *L. plantarum* in emulsion). Capital letters indicate significant differences between samples at the same storage time and different time; lowercase letters indicate significant differences between the same samples at different storage times.

over the storage period. The results were consistent with those reported by Bakirci and Kavaz [31], Singh and Muthukumarappan [32], Temiz et al. [28], and Tseng and Zhao [27]. Temiz et al. [28], Singh and Muthukumarappan [32], and Ertem and Cakmakci [33] also found that the acidity of yogurt increased during storage. The pH reduction and increase of titratable acidity during the storage time can be attributed to the activity of starter microorganisms. Encapsulation of probiotic bacteria had

considerable impacts on the acidity of samples. Free bacteria increased acidity more than encapsulated bacteria. Among probiotic bacteria, *L. rhamnosus* had a more significant effect on acidity. Similar results were reported by Chen et al. [15], who evaluated the effects of encapsulation of *Bifidobacterium* BB01 on the acidity of yogurt. In fact, the consumption of sugar and producing organic acids by microorganisms resulted in pH reduction and acidity increment [34].

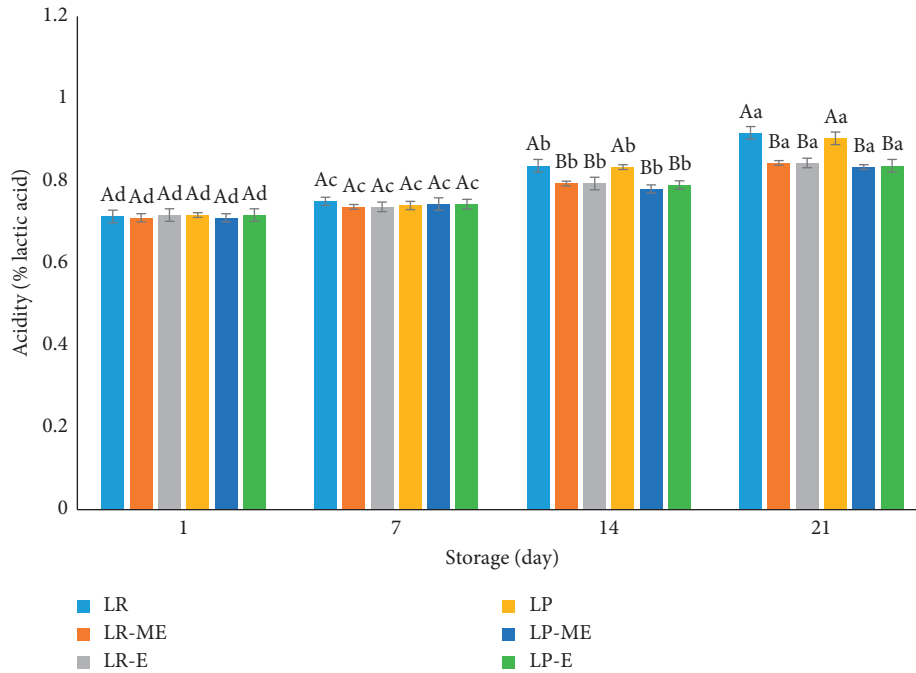


FIGURE 3: Acidity (% lactic acid) of probiotic yogurt. LR (*L. rhamnosus*), LR-ME (encapsulated *L. rhamnosus* in multilayer emulsion), LR-E (encapsulated *L. rhamnosus* in emulsion), LP (*L. plantarum*), LP-ME (encapsulated *L. plantarum* in multilayer emulsion), and LP-E (encapsulated *L. plantarum* in emulsion). Capital letters indicate significant differences between samples at the same storage time and different time; lowercase letters indicate significant differences between the same samples at different storage times.

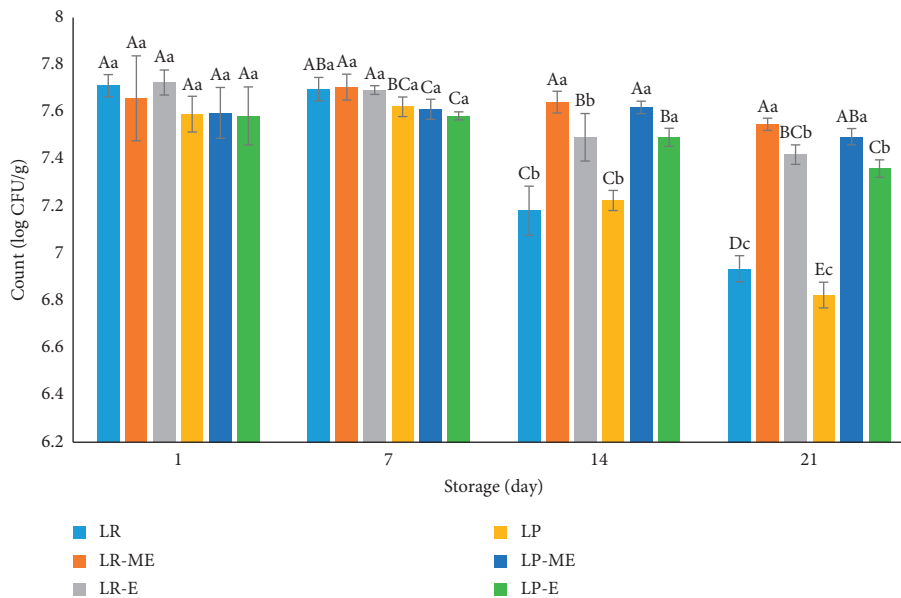


FIGURE 4: Viability of *L. rhamnosus* and *L. plantarum* of probiotic yogurt. LR (*L. rhamnosus*), LR-ME (encapsulated *L. rhamnosus* in multilayer emulsion), LR-E (encapsulated *L. rhamnosus* in emulsion), LP (*L. plantarum*), LP-ME (encapsulated *L. plantarum* in multilayer emulsion), and LP-E (encapsulated *L. plantarum* in emulsion). Capital letters indicate significant differences between samples at the same storage time and different time; lowercase letters indicate significant differences between the same samples at different storage times.

3.5. Viability of Probiotic and Starter Bacteria. Although there are many standards for the number of probiotic bacteria in yogurt, the acceptable number is usually reported as 10^7 CFU/g [35]. The number of probiotic bacteria was

enumerated every 7 days from the first day of production until 3 weeks. Figure 4 shows the number of *L. rhamnosus* and *L. plantarum* during the storage. As can be seen, the survival of bacteria has been significantly decreased during

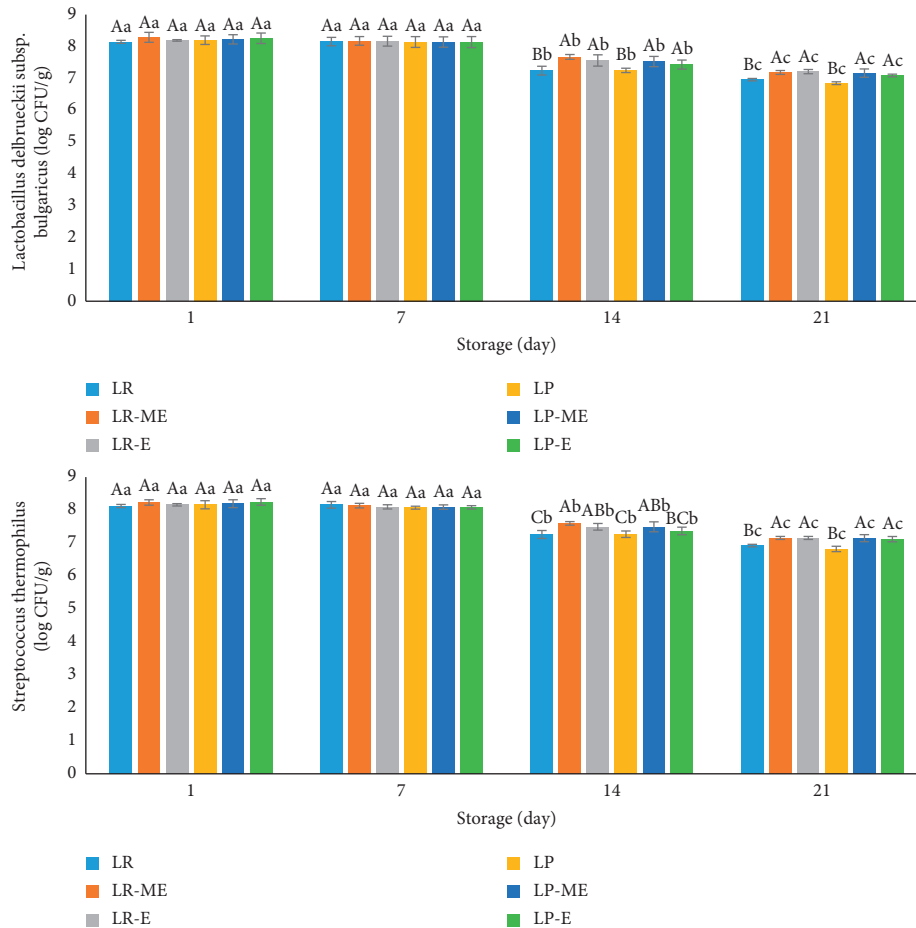


FIGURE 5: Starter count of probiotic yogurt. LR (*L. rhamnosus*), LR-ME (encapsulated *L. rhamnosus* in multilayer emulsion), LR-E (encapsulated *L. rhamnosus* in emulsion), LP (*L. plantarum*), LP-ME (encapsulated *L. plantarum* in multilayer emulsion), and LP-E (encapsulated *L. plantarum* in emulsion). Capital letters indicate significant differences between samples at the same storage time and different time; lowercase letters indicate significant differences between the same samples at different storage times.

the storage time. A similar observation was also reported by Eskandari et al. [36]. Encapsulation had a significant effect on the improving viability of probiotic bacteria, and samples encapsulated with multilayer emulsion had the highest viability. Encapsulated probiotic bacteria within emulsions can endure adverse conditions. These results are in agreement with those of Iqbal et al. [37] that showed survival of probiotic bacteria in yogurt. Also, Muzzafar and Sharma [14] reported the efficiency of different coating types on the survival of encapsulated bacteria compared to free bacteria in food products. Similarly, Qi et al. [38] declared higher survival of probiotics after encapsulation in yogurt. The number of encapsulated bacteria reduced by just 1.76 log CFU/g while the number of bacteria in the free form reduced significantly to 4.82 log CFU/g. Pradeep Prasanna and Charalampopoulos [39] also observed a considerable reduction of the number of free probiotics (3.67 log CFU/g) in comparison with the encapsulated form. According to Figure 4, the amount of survived population at the end of the storage period in encapsulated samples was more than 10^7 CFU/g. The results were consistent with those reported by Salwa et al. [40]. The results of the starter count were

reported in Figure 5. The bacteria count significantly reduced during storage time. This reduction was due to the consumption of the existing sugar by bacteria and the production of lactic acid. Also, after 14 days of storage, the samples containing free probiotic bacteria showed lower cell viability. The fact could be related to the lower level of pH.

3.6. Viscosity. Viscosity is one of the parameters that can be changed by the production of the acid components by different bacteria [41]. The effects of probiotic bacteria type (*L. rhamnosus* and *L. plantarum*) and encapsulation methods (simple and multilayer) on the viscosity of samples are presented in Figure 6. The results indicated that free bacteria decreased the viscosity of samples due to the production of acid components which affects the structure of samples. Our results were in accordance with those reported by Afzaal et al. [13] and Tarrega et al. [42]. The viscosity of LR, LR-ME, LR-E, LP, LP-ME, and LP-E samples were 65.86, 116.48, 88.97, 77.53, 124.56, and 93.25 mPs, respectively. After incorporating multilayer emulsion in the yogurt sample, the presence of protective layers around bacteria led to a lower

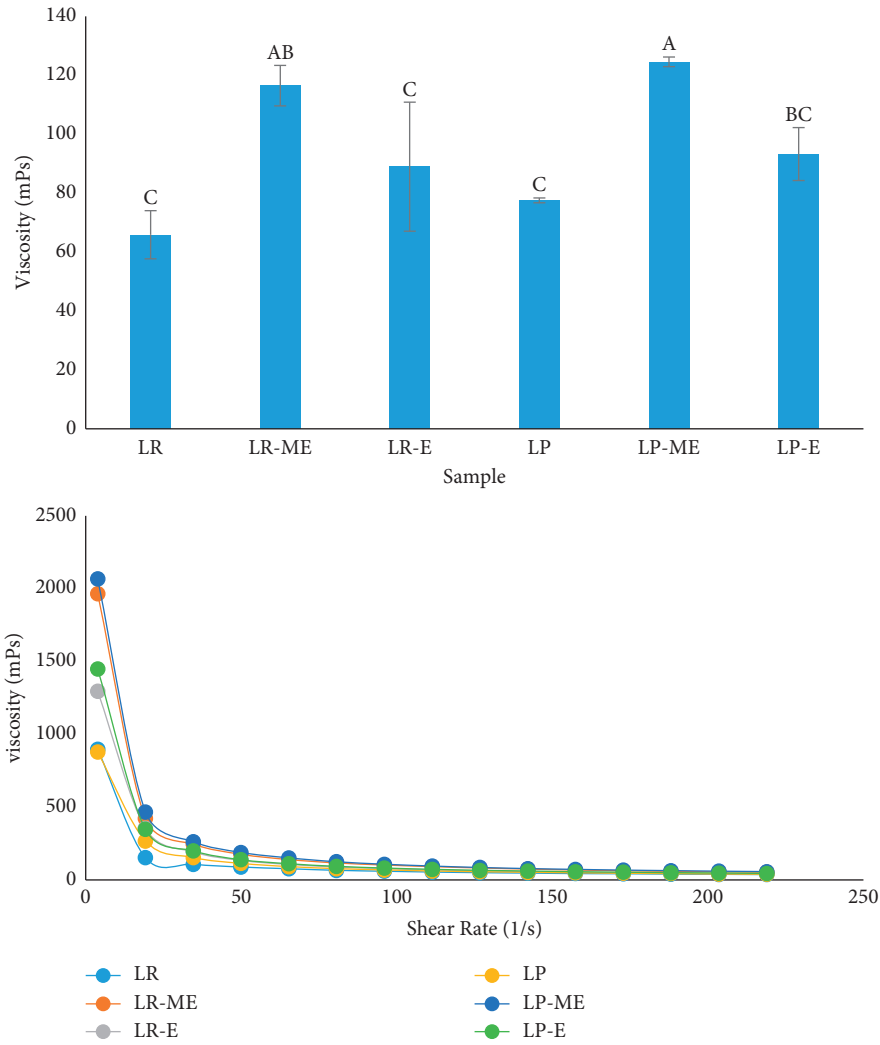


FIGURE 6: Viscosity of probiotic yogurt. LR (*L. rhamnosus*), LR-ME (encapsulated *L. rhamnosus* in multilayer emulsion), LR-E (encapsulated *L. rhamnosus* in emulsion), LP (*L. plantarum*), LP-ME (encapsulated *L. plantarum* in multilayer emulsion), and LP-E (encapsulated *L. plantarum* in emulsion).

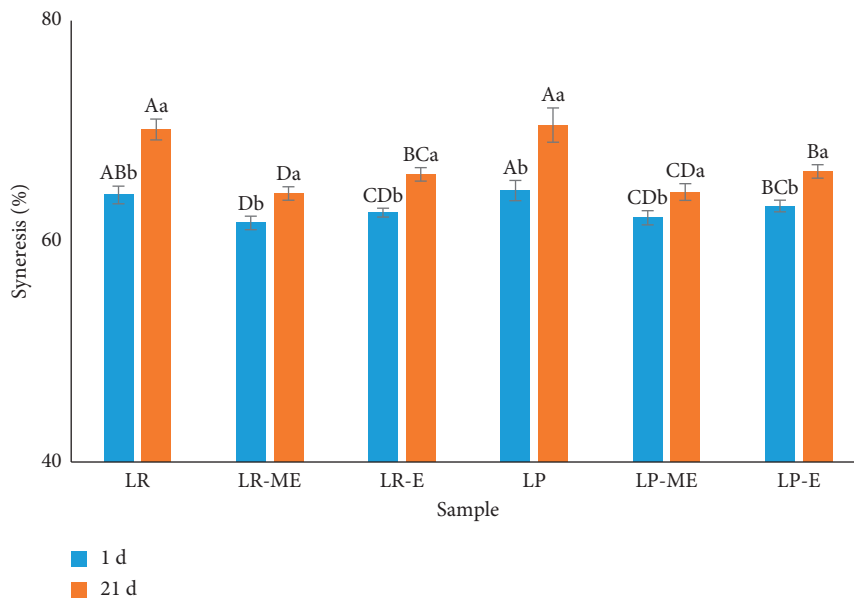


FIGURE 7: Syneresis of probiotic yogurt. LR (*L. rhamnosus*), LR-ME (encapsulated *L. rhamnosus* in multilayer emulsion), LR-E (encapsulated *L. rhamnosus* in emulsion), LP (*L. plantarum*), LP-ME (encapsulated *L. plantarum* in multilayer emulsion), and LP-E (encapsulated *L. plantarum* in emulsion). Capital letters indicate significant differences between samples at the same storage time and different time; lowercase letters indicate significant differences between the same samples at different storage times.

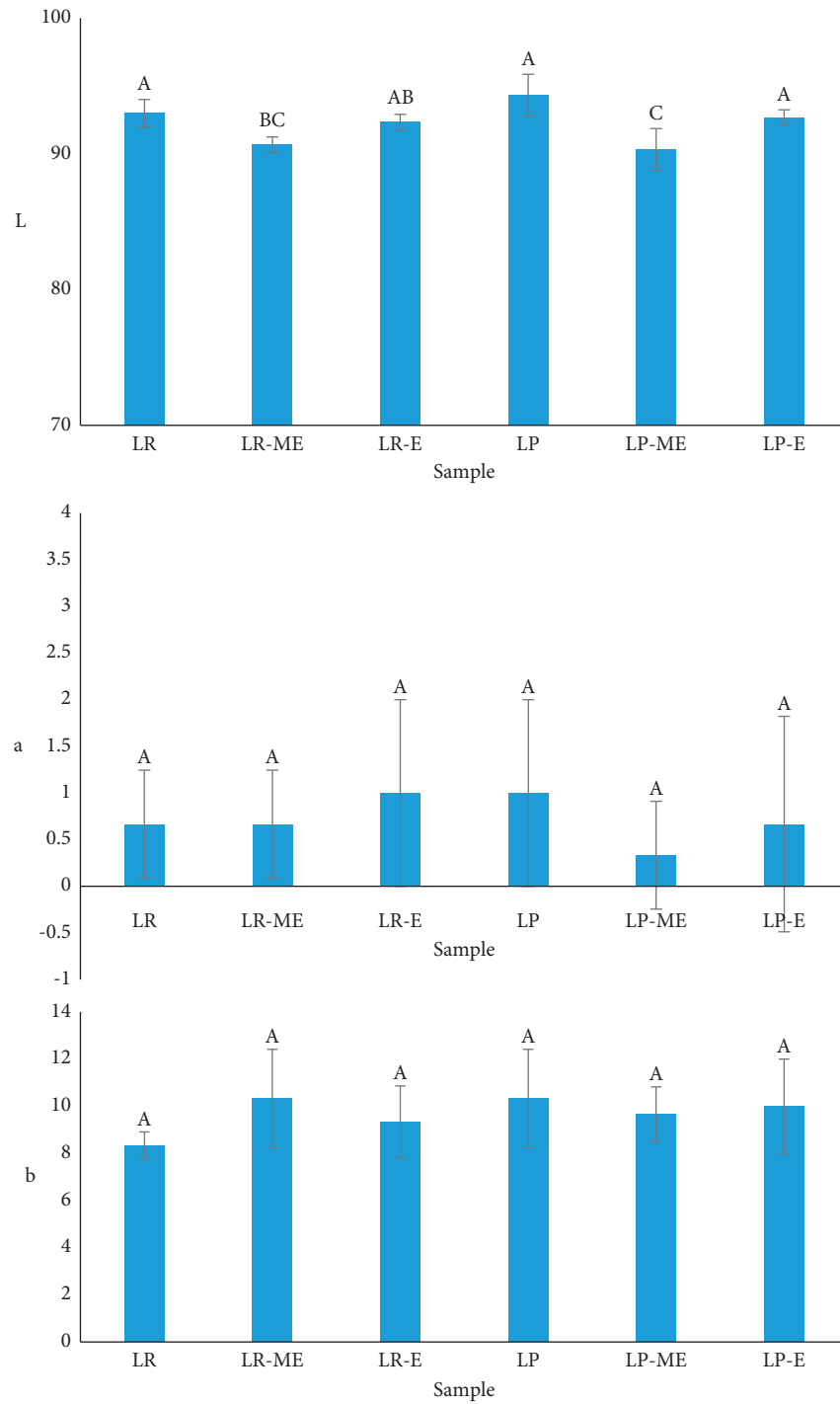


FIGURE 8: Color parameters of probiotic yogurt. LR (*L. rhamnosus*), LR-ME (encapsulated *L. rhamnosus* in multilayer emulsion), LR-E (encapsulated *L. rhamnosus* in emulsion), LP (*L. plantarum*), LP-ME (encapsulated *L. plantarum* in multilayer emulsion), and LP-E (encapsulated *L. plantarum* in emulsion).

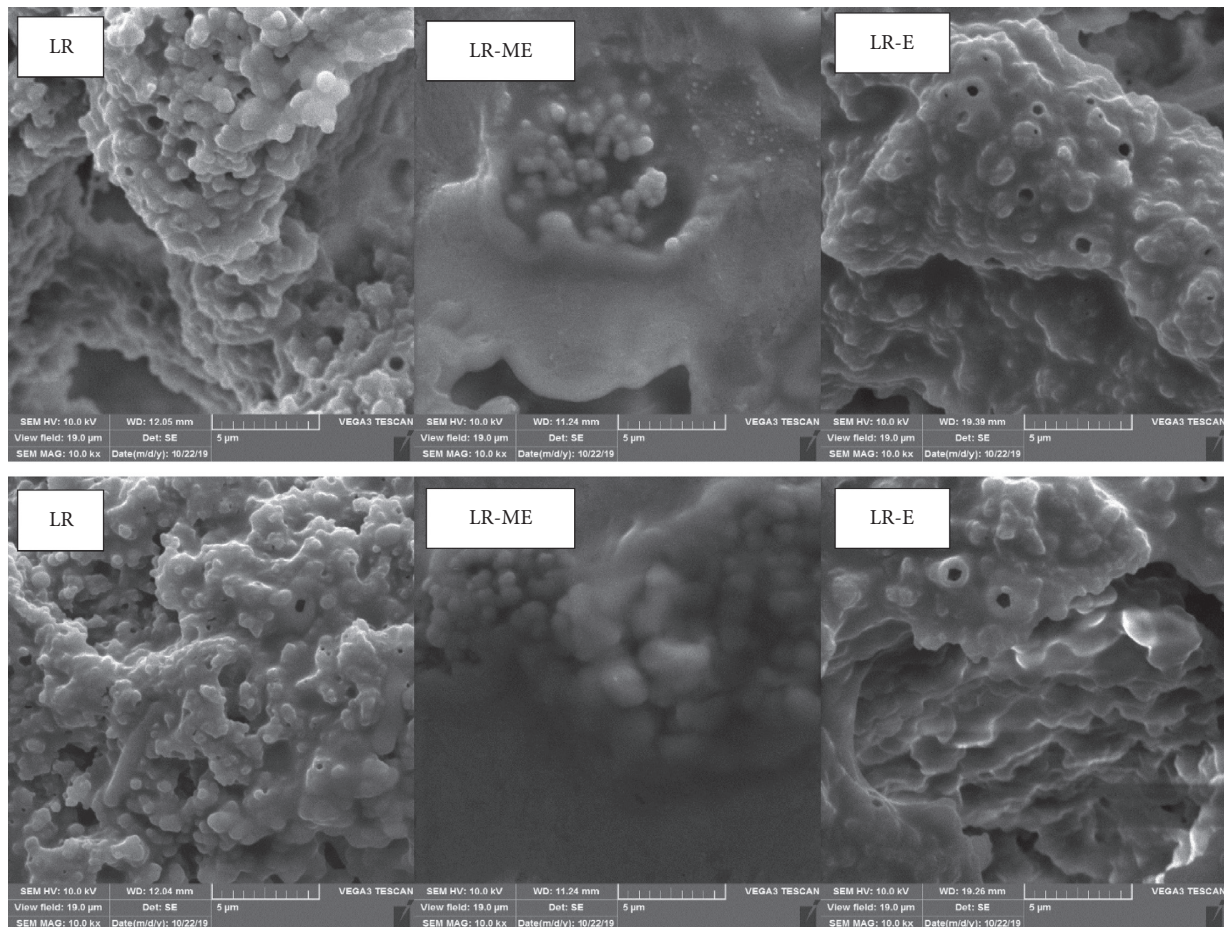


FIGURE 9: SEM micrograph parameters of probiotic yogurt. LR (*L. rhamnosus*), LR-ME (encapsulated *L. rhamnosus* in multilayer emulsion), LR-E (encapsulated *L. rhamnosus* in emulsion), LP (*L. plantarum*), LP-ME (encapsulated *L. plantarum* in multilayer emulsion), and LP-E (encapsulated *L. plantarum* in emulsion).

acidic components release. Moreover, Figure 6 shows the shear thinning behavior of all samples by increasing the shear rate.

3.7. Syneresis. The effects of probiotic bacteria type (*L. rhamnosus* and *L. plantarum*) and encapsulation methods (simple and multilayer) on the syneresis of samples are presented in Figure 7. It was indicated that free bacteria increased the syneresis of samples due to the production of high acid components. Moreover, the syneresis increment can be also related to the fact that the probiotics use solid materials and convert them into various metabolites. Similarly, Afzaal et al. [13] also reported higher syneresis value in yogurt after treatment with free probiotics compared to encapsulated ones.

3.8. Color. Color is one of the main properties of yogurt that can affect the consumer acceptability [43]. Figure 8 indicated

that probiotic bacteria type (*L. rhamnosus* and *L. plantarum*) and encapsulation methods (simple and multilayer) had no significant effects on the L^* , a^* , and b^* of samples.

3.9. SEM. The structure of the sample was evaluated by scanning electron microscopy. Figure 9 showed that probiotic bacteria types (*L. rhamnosus* and *L. plantarum*) had no significant effects on the structure of samples. However, samples incorporating multilayer emulsion were more homogenous due to the presence of WPI and Persian gum. Similar morphology was also reported by Fazilah et al. [44] and El Kadri et al. [45] after addition of double emulsion to yogurt samples.

3.10. Sensory Properties. Color, odor, texture, taste, strength, and overall acceptance of different yogurt samples were investigated by panelists. The effects of probiotic bacteria type (*L. rhamnosus* and *L. plantarum*) and encapsulation

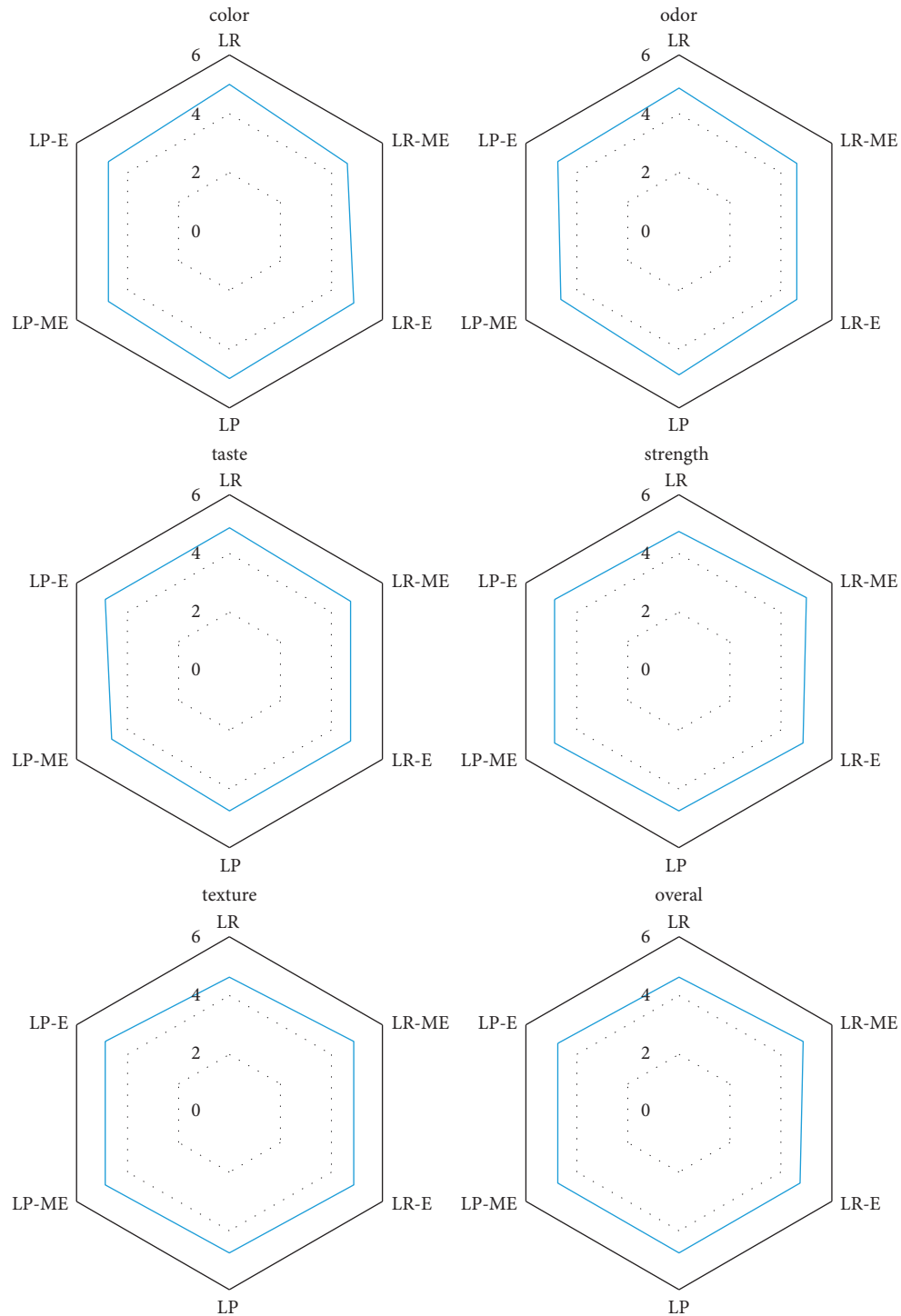


FIGURE 10: Sensorial properties parameters of probiotic yogurt. LR (*L. rhamnosus*), LR-ME (encapsulated *L. rhamnosus* in multilayer emulsion), LR-E (encapsulated *L. rhamnosus* in emulsion), LP (*L. plantarum*), LP-ME (encapsulated *L. plantarum* in multilayer emulsion), and LP-E (encapsulated *L. plantarum* in emulsion).

methods (simple and multilayer) on the sensorial aspects of yogurts are presented in Figure 10. It was revealed that probiotic bacteria type and encapsulation methods had no significant effects on the sensory properties of samples. Kailasapathy [46] also reported the constant sensorial properties of yogurt after encapsulation of probiotic bacteria.

4. Conclusion

It was observed that encapsulation of probiotics within both types of delivery systems enhanced the cell viability in yogurt samples. Sensorial characteristics and physicochemical parameters of the probiotic yogurts were also influenced by the encapsulation method. Based on the results, multilayer

emulsion was an effective tool to preserve the viability of bacteria at the recommended effectiveness level (above 10^7 CFU/g) in food products with low negative effects on the physicochemical and sensorial properties.

Data Availability

The data used to support this study are available from the corresponding author upon request.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

- [1] J. I. Boye, *Nutraceutical and Functional Food Processing Technology*, John Wiley & Sons, New York, NY, USA, 2015.
- [2] S. Gupta, J. J. Lee, and W. N. Chen, "Analysis of improved nutritional composition of potential functional food (Okara) after probiotic solid-state fermentation," *Journal of Agricultural and Food Chemistry*, vol. 66, no. 21, pp. 5373–5381, 2018.
- [3] P. Roy and V. Kumar, "Functional food: probiotic as health booster," *Journal of Food, Nutrition and Population Health*, vol. 2, no. 2, pp. 12–15, 2018.
- [4] Z. S. Ladjevardi, M. Yarmand, Z. Emam-Djomeh, and A. Niasari-Naslaji, "Physicochemical properties and viability of probiotic bacteria of functional synbiotic camel yogurt affected by oat β -glucan during storage," *Journal of Agriculture, Science and Technology*, vol. 18, pp. 1233–1246, 2018.
- [5] S. Sarkar, "Potentiality of probiotic yoghurt as a functional food—a review," *Nutrition & Food Science*, vol. 49, no. 2, pp. 182–202, 2019.
- [6] R. Karimi, A. M. Mortazavian, and A. G. Da Cruz, "Viability of probiotic microorganisms in cheese during production and storage: a review," *Dairy Science & Technology*, vol. 91, no. 3, pp. 283–308, 2011.
- [7] K. WoldemariamYohannes, Z. Wan, Q. Yu et al., "Prebiotic, Probiotic, antimicrobial, and functional food applications of *Bacillus amyloliquefaciens*," *Journal of Agricultural and Food Chemistry*, vol. 68, no. 50, pp. 14709–14727, 2020.
- [8] S. Mishra and H. Mishra, "Effect of synbiotic interaction of fructooligosaccharide and probiotics on the acidification profile, textural and rheological characteristics of fermented soy milk," *Food and Bioprocess Technology*, vol. 6, no. 11, pp. 3166–3176, 2013.
- [9] M. Shakibaie, T. Mohammadi-Khorsand, M. Adeli-Sardou et al., "Probiotic and antioxidant properties of selenium-enriched *Lactobacillus brevis* LSe isolated from an Iranian traditional dairy product," *Journal of Trace Elements in Medicine & Biology*, vol. 40, pp. 1–9, 2017.
- [10] S. Hasani, I. Khodadadi, and A. Heshmati, "Viability of *Lactobacillus acidophilus* in rice bran-enriched stirred yoghurt and the physicochemical and sensory characteristics of product during refrigerated storage," *International Journal of Food Science and Technology*, vol. 51, no. 11, pp. 2485–2492, 2016.
- [11] R. Ferdousi, M. Rouhi, R. Mohammadi, A. M. Mortazavian, K. Khosravi-Darani, and A. H. Rad, "Evaluation of probiotic survivability in yogurt exposed to cold chain interruption," *Iranian Journal of Pharmaceutical Research: Iranian Journal of Pharmaceutical Research*, vol. 12, 2013.
- [12] J. T. Guimarães, C. F. Balthazar, H. Scudino et al., "High-intensity ultrasound: a novel technology for the development of probiotic and prebiotic dairy products," *Ultrasonics Sonochemistry*, vol. 57, pp. 12–21, 2019.
- [13] M. Afzaal, A. U. Khan, F. Saeed et al., "Functional exploration of free and encapsulated probiotic bacteria in yogurt and simulated gastrointestinal conditions," *Food Sciences and Nutrition*, vol. 7, no. 12, pp. 3931–3940, 2019.
- [14] A. Muzzafar and V. Sharma, "Microencapsulation of probiotics for incorporation in cream biscuits," *Journal of Food Measurement and Characterization*, vol. 12, no. 3, pp. 2193–2201, 2018.
- [15] L. Chen, T. Yang, Y. Song, G. Shu, and H. Chen, "Effect of xanthan-chitosan-xanthan double layer encapsulation on survival of Bifidobacterium BB01 in simulated gastrointestinal conditions, bile salt solution and yogurt," *LWT-Food Science and Technology*, vol. 81, pp. 274–280, 2017.
- [16] N. F. Fazilah, A. B. Ariff, M. E. Khayat, L. Rios-Solis, and M. Halim, "Influence of probiotics, prebiotics, synbiotics and bioactive phytochemicals on the formulation of functional yogurt," *Journal of Functional Foods*, vol. 48, pp. 387–399, 2018.
- [17] L. Zhao, R. Feng, F. Ren, and X. Mao, "Addition of buttermilk improves the flavor and volatile compound profiles of low-fat yogurt," *Lebensmittel-Wissenschaft & Technologie*, vol. 98, pp. 9–17, 2018.
- [18] T. A. Comunian, I. E. Chaves, M. Thomazini et al., "Development of functional yogurt containing free and encapsulated echium oil, phytosterol and sinapic acid," *Food Chemistry*, vol. 237, pp. 948–956, 2017.
- [19] M. Colombo, S. D. Todorov, M. Eller, and L. A. Nero, "The potential use of probiotic and beneficial bacteria in the Brazilian dairy industry," *Journal of Dairy Research*, vol. 85, no. 4, pp. 487–496, 2018.
- [20] H. H. Gahrue, M. H. Eskandari, and G. Mesbahi, "Development of functional yogurt fortified with wheat germ and strawberry as functional ingredients," *Progress in Nutrition*, vol. 21, pp. 388–398, 2019.
- [21] S. M. H. Hosseini, H. H. Gahrue, M. Razmjooie et al., "Effects of novel and conventional thermal treatments on the physicochemical properties of iron-loaded double emulsions," *Food Chemistry*, vol. 270, pp. 70–77, 2019.
- [22] A. Gilbert, L. E. Rioux, D. St-Gelais, and S. L. Turgeon, "Studying stirred yogurt microstructure using optical microscopy: how smoothing temperature and storage time affect microgel size related to syneresis," *Journal of Dairy Science*, vol. 103, no. 3, pp. 2139–2152, 2020.
- [23] I. Ścibisz, M. Ziarno, and M. Mitek, "Color stability of fruit yogurt during storage," *Journal of Food Science & Technology*, vol. 56, no. 4, pp. 1997–2009, 2019.
- [24] A. R. Karnopp, K. G. Oliveira, E. F. de Andrade, B. M. Postinger, and D. Granato, "Optimization of an organic yogurt based on sensorial, nutritional, and functional perspectives," *Food Chemistry*, vol. 233, pp. 401–411, 2017.
- [25] D. W. Ningtyas, B. Bhandari, N. Bansal, and S. Prakash, "The viability of probiotic *Lactobacillus rhamnosus* (non-encapsulated and encapsulated) in functional reduced-fat cream cheese and its textural properties during storage," *Food Control*, vol. 100, pp. 8–16, 2019.
- [26] C. S. Ranadheera, C. Evans, M. Adams, and S. Baines, "Probiotic viability and physico-chemical and sensory properties of plain and stirred fruit yogurts made from goat's milk," *Food Chemistry*, vol. 135, no. 3, pp. 1411–1418, 2012.

- [27] A. Tseng and Y. Zhao, "Wine grape pomace as antioxidant dietary fibre for enhancing nutritional value and improving storability of yogurt and salad dressing," *Food Chemistry*, vol. 138, no. 1, pp. 356–365, 2013.
- [28] H. Temiz, Z. Tarakci, T. Karadeniz, and T. Bak, "The effect of loquat fruit (*Eriobotrya japonica*) marmalade addition and storage time on physico-chemical and sensory properties of yogurt," *Journal of Agricultural Sciences*, vol. 18, pp. 329–338, 2012.
- [29] T. Turgut and S. Cakmakci, "Probiotic strawberry yogurts: microbiological, chemical and sensory properties," *Probiotics & Antimicrobial Proteins*, vol. 10, pp. 64–70, 2018.
- [30] P. Panesar and C. Shinde, "Effect of storage on syneresis, pH, *Lactobacillus acidophilus* count, Bifidobacterium bifidum count of Aloe vera fortified probiotic yoghurt," *Current Research in Dairy Sciences*, vol. 4, 2012.
- [31] I. Bakirci and A. Kavaz, "An investigation of some properties of banana yogurts made with commercial ABT-2 starter culture during storage," *International Journal of Dairy Technology*, vol. 61, no. 3, pp. 270–276, 2008.
- [32] G. Singh and K. Muthukumarappan, "Influence of calcium fortification on sensory, physical and rheological characteristics of fruit yogurt," *LWT-Food Science and Technology*, vol. 41, no. 7, pp. 1145–1152, 2008.
- [33] H. Ertem and S. Cakmakci, "Shelf life and quality of probiotic yogurt produced with *Lactobacillus acidophilus* and Gobdin," *International Journal of Food Science and Technology*, vol. 53, pp. 776–783, 2018.
- [34] H. H. Gahruie, M. H. Eskandari, G. Mesbahi, and M. A. Hanifpour, "Scientific and technical aspects of yogurt fortification: a review," *Food Science and Human Wellness*, vol. 4, no. 1, pp. 1–8, 2015.
- [35] C. Fadela, C. Abderrahim, and B. Ahmed, "Sensorial and physico-chemical characteristics of yoghurt manufactured with Ewe's and skim milk," *World Journal of Dairy & Food Sciences*, vol. 4, no. 2, pp. 136–140, 2009.
- [36] M. H. Eskandari, A. Baroutkoub, M. Roushan Zamir, R. Beglarian, I. Ghasemkhani, and S. Shekarforoush, "Effect of milk supplementation on growth and viability of starter and probiotic bacteria in yogurt during refrigerated storage," *Iranian Journal of Veterinary Research*, vol. 13, no. 3, pp. 195–202, 2012.
- [37] R. Iqbal, T. Zahoor, N. Huma, A. Jamil, and G. Ünlü, "In-vitro GIT tolerance of microencapsulated *Bifidobacterium bifidum* ATCC 35914 using polysaccharide-protein matrix," *Probiotics and Antimicrobial Proteins*, vol. 11, no. 3, pp. 830–839, 2019.
- [38] W. Qi, X. Liang, T. Yun, and W. Guo, "Growth and survival of microencapsulated probiotics prepared by emulsion and internal gelation," *Journal of Food Science & Technology*, vol. 56, no. 3, pp. 1398–1404, 2019.
- [39] P. Pradeep Prasanna and D. Charalampopoulos, "Encapsulation in an alginate-goats' milk-inulin matrix improves survival of probiotic Bifidobacterium in simulated gastrointestinal conditions and goats' milk yoghurt," *International Journal of Dairy Technology*, vol. 72, no. 1, pp. 132–141, 2019.
- [40] A. A. Salwa, E. Galal, and A. E. Neimat, "Carrot yoghurt: sensory, chemical, microbiological properties and consumer acceptance," *Pakistan Journal of Nutrition*, vol. 3, no. 6, pp. 322–330, 2004.
- [41] Z. Ghasempour, N. Javanmard, A. M. Langroodi, M. Alizadeh-Sani, A. Ehsani, and E. M. Kia, "Development of probiotic yogurt containing red beet extract and basil seed gum; techno-functional, microbial and sensorial characterization," *Biocatalysis and Agricultural Biotechnology*, vol. 29, Article ID 101785, 2020.
- [42] A. Tarrega, J. Marcano, and S. Fiszman, "Yogurt viscosity and fruit pieces affect satiating capacity expectations," *Food Research International*, vol. 89, pp. 574–581, 2016.
- [43] A. Aghajani, S. A. Mortazav, F. T. Yazdi, M. S. Zenosian, and M. R. S. Asl, "Color, microbiological and sensory properties of low-fat probiotic yogurt supplemented with *Spirulina platensis* and *Ferulago angulata* hydroalcoholic extracts during cold storage," *Banat's Journal of Biotechnology*, vol. 10, no. 19, pp. 20–34, 2019.
- [44] N. Fazilah, N. Zani, H. Wasoh, A. Ariff, and M. Halim, "Influence of miracle fruit (*Synsepalum dulcificum*) extract and microencapsulated *Lactococcus lactis* Gh1 on the antioxidant activity and probiotic viability of functional yogurt," *International Food Research Journal*, vol. 27, no. 4, 2020.
- [45] H. El Kadri, S. Lalou, F. Mantzouridou, and K. Gkatzionis, "Utilisation of water-in-oil-water ($W_1/O/W_2$) double emulsion in a set-type yogurt model for the delivery of probiotic *Lactobacillus paracasei*," *Food Research International*, vol. 107, pp. 325–336, 2018.
- [46] K. Kailasapathy, "Survival of free and encapsulated probiotic bacteria and their effect on the sensory properties of yoghurt," *LWT-Food Science and Technology*, vol. 39, no. 10, pp. 1221–1227, 2006.