Boosting effects of *Spirulina platensis*, whey protein, and probiotics on the growth of microflora and the nutritional value of ayran

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Abstract
Great attention has been given to *Spirulina platensis* for consideration as the superfood of the future due to its high bioactive compounds and nutritional values. Boosting effects of *S. platensis* and whey protein hydrolysates (WPH) on the growth of probiotics in ayran, a fermented milk product, through the fermentation and storage periods were investigated in this study. This study aimed to enhance the growth of *Lactobacillus delbrueckii* subsp. *bulgaricus*, *Bifidobacterium lactis*, *Streptococcus thermophilus*, and *L. acidophilus* with the addition of *S. platensis* and WPH added or not probiotics into ayran. Effects of *S. platensis* and WPH (at 0%, 0.25%, 0.5%, and 1%) on the growth of probiotic culture were studied before and after the fermentation and at seventh, 14th, and 21st days of storage. *S. platensis* and WPH had significant boosting effects (*P* < .05) on the growth of *St. thermophilus* (9.22 log cfu/mL), *L. delbrueckii* subsp. *bulgaricus* (6.85 log cfu/mL), *L. acidophilus* (5.39 log cfu/mL), and *B. lactis* (7.96 log cfu/mL) and biochemical variables. The highest total solid level (0.99) and protein content (25.16 mg/L) were observed by mixing of *S. platensis* and WPH at 1% into ayran. The encountered variability in the boosting effect of *S. platensis* and WPH depended on their addition to ayran. *S. platensis* and WPH had great potentials for enhancing the nutritional value of ayran and the growth of the probiotic culture.

KEYWORDS
ayran, fermentation, probiotic culture, *Spirulina platensis*, whey protein hydrolysates

## 1 | INTRODUCTION

Fermented dairy products such as kefir, ayran, yoghurt, and so on are nutrient-rich foods that are considered to be one of the most popular fermented milk products around the world.¹,² The general utilization of milk products, especially, probiotic-milk products provide an excellent assessment of long-term foods in health with the natural food and improve the intestinal flora with existing lactic acid bacteria.³ Probiotic bacteria are related to various health advantages and the most important for probiotic is viability and can survive in the gastrointestinal tract at a specific number, improve the...
microbial balance of the intestinal system and stay survive in the different conditions. The increment in demand for a healthy diet requires innovation and new product development in food manufacture widely. The food manufacturer has a great role in providing healthier foods and assisting healthier eating habits.

The addition of *Spirulina platensis* (a protein-rich blue-green alga) on the viability of beneficial culture in yoghurt is rather crucial for promoting the growth of probiotics. *S. platensis* has a positive influence on the growth of *Lactobacillus* in the intestinal system and promote acid production of *Lactococci*. Previous studies reported that the influence of *S. platensis* can improve the starter culture growth and viability in probiotic type yoghurt.

The production of fermented or unfermented whey protein hydrolysates (WPH) can be used to improve the activity of probiotics and support health-promoting dairy products. These types of drinks, alternative for probiotics, WPH progress the ability of lactic acid bacteria because it works for promoting the perfect viability of fermented milk products. Besides, the addition of *S. platensis* into probiotic-fermented dairy products will enhance the ability of probiotics and their functional characteristics because this cyanobacterium contains a huge quantity of nutrients taken into account “a functional food”.

Hence, the idea of combining *S. platensis* with WPH which can be used as dietary supplements in dairy products to achieve high efficiency in increasing the growth of probiotics. The nutritional importance of the mixed form of *S. platensis*, WPH, and probiotics can be considered highly nutritious and cost-effective due to containing a senior count of probiotic bacteria. The present study aimed to enhance the growth of probiotic culture (*Lactobacillus delbrueckii* subsp. *bulgaricus*, *L. acidophilus*, *Bifidobacterium lactis*, and *Streptococcus thermophilus*) after adding *S. platensis* and WPH added or not probiotics into ayran before and after the fermentation and the storage period. Besides, the addition of these supplements into ayran not only promotes the ability of probiotic bacteria but also enhance nutritional and physico-chemical levels, which can make a functional food.

## MATERIALS AND METHODS

### 2.1 Experimental design

The experimental design is given in Figure 1. Fresh milk was pasteurized for 15 minutes at 95°C. After cooling to 40°C, yoghurt starter (4% v/v), powder of *S. platensis* (Cyanotech Corp., Hawaii, USA) and WPH at 0 (the control), 0.25%, 0.5%, and 1.0% (w/v) concentrations added or not probiotics (*Enterococcus faecium*, *L. acidophilus*, *L. rhamnosus*, *B. lactis*, and *B. bifidum* contain $2.5 \times 10^9$ cfu/2 g) were incorporated into pasteurized milk. A Probiotics sachet (2 g) was dissolved in 100 mL of sterilized peptone water at room temperature, hold for 30 minutes and added as 0.25%, 0.5%, and 1% (v/v). Thereafter, 0.5% salt (w/v) and 50% sterile water (v/v) were mixed. After that, each mixture was incubated at 40°C for fermentation. The fermentation process carried out up to pH value reach to about 4.4 (about 4 hours). Each of the samples was cooled down to 4°C after the fermentation, and they were held at 4 ± 1°C for 21 days. As a physico-chemical property such as the acidity and pH values of each sample and the counting of probiotic culture were followed through

![FIGURE 1 Experimental design](image)
the fermentation period, after the fermentation and within the storage time at seventh, 14th and 21st days. Hunter color and viscosity measurements were done after 1 day of sampling.

2.2 Microbiological analysis

Counting of bacteria (log cfu/mL) in ayran having different concentrations of \textit{S. platensis} and WPH adding or not probiotics were made before and after the fermentation and storage time. Tenfold diluted the samples with peptone water (0.1% v/v) were poured or spread to agar after than Petri dishes were incubated. After that, the colonies of bacteria were enumerated on duplicated Petri plates according to the method proposed by Dave and Shah.\textsuperscript{17} Petri plates were incubated anaerobically at 5% CO\textsubscript{2}. Counting of \textit{L. acidophilus} was done by the spread plate technique on to MRS agar with adding 1% of maltose contained inoculation of appropriate dilution. Petri plates were incubated at 37°C for 3 days, and then the viable colonies were enumerated. Colonies of \textit{St. thermophilus} were counted by the inoculation with the pour plate method on to M17 agar containing 1% of lactose. Petri dishes were held for 24 hours at 45°C and colonies were counted. Viable \textit{L. delbrueckii ssp. bulgaricus} colonies were counted into MRS agar with 1% of fructose by the pour-plate technique and then Petri dishes were incubated for 3 days at 45°C and counted. Enumeration of \textit{B. lactis} colonies on the appropriate dilutions into MRS agar was carried out by the pour plate method. Petri plates were incubated for 3 days at 45°C and the colonies were counted.

2.3 Physico-chemical analyses

Values of pH were measured using a pH meter every 1 hour during the fermentation of ayran samples. Each sample was titrated with 0.1M NaOH by the use of phenolphthalein indicator for recording the value of acidity. The value of NaOH which obtained as a result of calibration used multiplied by 2 and multiplied by 0.0225 as a fixed value. Soxhlet-Henkel point value was a percentage of acidity. The titratable acidity was determined before and after the fermentation and during the storage period.

Total solid values of samples were quantified by the use of an oven method reported in the TS1330.\textsuperscript{16} Briefly, the samples were put into a beaker in a boiling water bath up to remove the excess water from the samples and then were cooled at room temperature. The dishes were heated in an oven for 1 hour at 102°C to 103°C and then the dishes were cooled down in a desiccator. After then their weight was recorded as (\(M_0\)). The 5 mL of sample was added and recorded the weight before drying (\(M_1\)), then put it in the oven at 102°C to 103°C up to constant weight reached. The dishes were removed and put in a desiccator to arrive at room temperature and recorded the weight after drying (\(M_2\)). The total solids of samples were determined by an Equation (1).

\[
\text{%Total solids} = \frac{(M_2 - M_0)}{(M_1 - M_0)} \times 100.
\]  

Protein contents of samples were determined according to the Turkish standard method TS1330.\textsuperscript{16} Ten milliliter of sample, 0.4 mL saturated potassium oxalate and 0.5 mL of phenolphthalein solution were mixed very well in a flask and held for 2 minutes. Thereafter, 0.1M NaOH solution in the burette was used for the neutralization of the mixture until getting a faint color. Then, a 40% formaldehyde solution neutralized with phenolphthalein and 0.1M sodium hydroxide solution was added as 2 mL into each sample mixtures. And then, each of the samples was titrated till getting the same pink color as the previous color to determine the level of 0.1M sodium hydroxide used for the second titration. The protein content of milk is determined by Equations (2) and (3).

\[
\text{%Total protein} = \text{Aldehyde value} \times 0.17, \quad (2)
\]

\[
\text{%Total protein} = 1.7 \times (a - b), \quad (3)
\]

where \(a\) is the used NaOH (milk) as mL, and \(b\) is the used NaOH as mL for blank (water).

Viscosity values of samples were determined after 1 day of sampling time taking at 7, 14, 21 days of storage by the use of a viscometer (Brookfield viscometer, DV3T, USA) in 150 mL beaker at 20 rpm.
The colors of ayran samples were read after the fermentation period and the storage period by the use of Hunter color equipment (Hunter Lab ColorFlex, Reston, VA). About 10 mL of samples were taken in a special class and put on the equipment, and then recorded the measurements.

2.4 | Statistical analyses

Statistical analyses were carried out to determine the effects of *S. platensis* and WHP added or not probiotics and time on the growth of probiotic culture and other parameters. Analysis of variance was applied to compare the mean of, colors, viscosity, number of probiotic cultures, pH, and titratable acidity obtained from different conditions at $\alpha = .05$ level, by use of SPSS version 19.0 (SPSS Inc., Chicago, USA). Duncan’s multiple range test was carried out for comparing more than two sample groups.

3 | RESULTS AND DISCUSSION

3.1 | Growth of *Streptococcus thermophilus*

Effects of *S. platensis* + WPH added or not probiotics on the viable count of *St. thermophilus* in the ayran were monitored before and after fermentation and during the storage time (Figure 2A). The highest growth of *St. thermophilus* (9.22 log cfu) was seen in the sample with the addition of 0.25% mixture of *S. platensis* + WPH + probiotics at 21st days of storage. Results indicated that viable *St. thermophilus* in the present study was higher than a previous study related to the probiotic bacteria in the algal yoghurt. Comparing to the control sample, mixing of *S. platensis* and WPH with probiotics showed significant ($P < .05$) effects on the growth of *St. thermophilus* before fermentation. The increase in *St. thermophilus* growth could be due to the high concentration of nutrients in *S. platensis* and WPH such as minerals, vitamins (especially vitamin B), and amino acids. It is known that nutrients are a promoter for the growth of probiotic culture.

3.2 | Growth of *Lactobacillus delbrueckii* subsp. *bulgaricus*

The growth of *L. delbrueckii* subsp. *bulgaricus* in each sample was monitored before and after the fermentation and within the storage time (Figure 2B). Compared with the control group, the supplement of *S. platensis* + WPH mixtures at different percentages did not enhance *L. bulgaricus* growth ($P > .05$) up to 14 days of storage. The mixing of *S. platensis* + WPH + probiotics had significant ($P < .05$) boosting effects on the growth of *L. bulgaricus* after the fermentation and at 14th, and 21st days of storage. This could be due to the synergic effects of biochemical compounds in this mixture. Varga et al. expressed that *S. platensis* contain nitrogenous compounds such as peptone and free amino acids that have improving effects on the growth of *L. bulgaricus*. Martelli et al. reported that the boosting effect of *S. platensis* on lactic acid bacteria strains was significantly higher than that of yeast extract. The existence of hypoxanthine, free amino acids, peptone, and adenine in *Spirulina* biomass increased the viable count of *L. bulgaricus*. The addition of whey protein at 1% and honey at 2% and 4% improved the growth of *L. bulgaricus* in the yoghurt through 21st of storage.

3.3 | Growth of *Lactobacillus acidophilus*

Effects of *S. platensis* + WPH added or not probiotics on the growth of *L. acidophilus* in the ayran were determined before and after the fermentation and during the storage (Figure 3A). The highest viable count of *L. acidophilus* was found in the samples containing 1% of the mixture of *S. platensis* + WPH (5.39 log cfu) at 21st days of storage. Increase in additive supplements enhanced ($P < .05$) the growth of *L. acidophilus*. As reported by a previous study, nutrients are required for the growth and survival of *L. acidophilus*. These supplements especially *S. platensis* powder is a unique source of nutrients for these bacteria since it contains a significant amount of essential amino acids, vitamins, mineral, and so on. In addition, Fox reported that the derivate of vitamin B is an important promoter for probiotic bacteria. Bhownik et al.
observed that *S. platensis* promoted the growth of probiotics. The presence of nutrients in *S. platensis* biomass such as free amino acids, hypoxanthine, minerals, exopolysaccharide, adenine, and vitamins had a positive influence on promoting the growth of probiotics.\(^5\)

### 3.4 Growth of *Bifidobacterium lactis*

Effects of *S. platensis* + WPH added or not probiotics on the growth of *B. lactis* are given in Figure 3B. The highest growth of *B. lactis* was recorded in the sample containing 0.5% *S. platensis* + WPH + probiotics as 7.96 log cfu/mL. It can be stated that *S. platensis* improved the survival of the *Bifidobacterium* in the storage period of ayran.\(^{13}\) Akalin et al.\(^{14}\) reported that the growth of *B. animalis* increased when WPH added in low-fat yoghurt and full-fat milk compared with the control group through the first week of storage. This could be due to the increasing concentration of amino acids by WPH considered as a nutrient for probiotic culture. Similar results were found in the growth of probiotics in milk supplements by adding WPH improved the growth of *B. longum*.\(^{25}\)
FIGURE 3  Effect of addition of *Spirulina platensis* and WPH added or not probiotics on the growth of (A) *Lactobacillus acidophilus* and (B) *Bifidobacterium lactis* through fermentation and storage time. SW is *Spirulina platensis* + WPH. SWP is *Spirulina platensis* + WPH + probiotics. All the points are the mean of four data. WPH, whey protein hydrolysates.

### 3.5 Value of pH and titratable acidity

The effects of *S. platensis*, WPH, and probiotics incorporation on the pH levels of samples were monitored during the study period and their results are shown in Table 1. Statistical analyses indicated that an increase in these supplement amounts showed a significant (*P* < .05) boosting effect on the pH value compared with the control group after the fermentation and during the storage period (Table 1). Due to the addition of some biomolecules with high isoelectric points could increase in pH value. The samples prepared with *S. platensis* appeared to have higher buffering activity may that reason for the enrichment of ayan by amino acids, peptides, and proteins. An increase in the buffering capacity caused to decrease in pH value slowly due to that they prevent fermentation extremely. The addition of WPH can increase lactic acid production and this reduced pH level. They also recorded that WPH may be able to be used to control the pH levels.

The titratable acidity levels of each sample were followed before fermentation, after the fermentation and during the storage time at the different values of *S. platensis* + WPH added or not probiotics (0.25%, 0.5%, and 1%). Duncan’s multiple range tests showed that supplements significantly affect (*P* < .05) titratable acidity levels of samples (Table 1). The lowest titratable acidity was found in the sample contain 0.25% mixture of *S. platensis* and WPH at the end of 21 days. Samples containing 0.25% mixture of *S. platensis* and WPH decreased titratable acidity compared with the control (*P* < .05) and
Table 1: Effect of addition of *Spirulina platensis* and WPH with/without probiotic on the pH and acidity (Soxhlet Henkel) values through the fermentation and storage time

| Sample                | %  | Before fermentation | After fermentation | After 7 days   | After 14 days  | After 21 days  |
|-----------------------|----|---------------------|--------------------|----------------|----------------|---------------|
| Control               | 0  | 6.14a ± 0.29        | 4.69d ± 0.29       | 4.33c ± 0.29   | 4.06a ± 0.294  | 4.13b ± 0.29  |
| *Spirulina* + WPH     | 0.25| 6.35b ± 0.32        | 4.33d ± 0.32       | 4.12a ± 0.32   | 4.16b ± 0.3    | 4.23bc ± 0.32 |
|                       | 0  | 6.65c ± 0.34        | 4.50d ± 0.34       | 4.27c ± 0.34   | 4.23a ± 0.3    | 4.35bc ± 0.34 |
| *Spirulina* + WPH + Probiotic | 0.25| 6.35b ± 0.31        | 4.32d ± 0.31       | 4.15c ± 0.31   | 4.03a ± 0.31   | 4.27bc ± 0.31 |
|                       | 0  | 6.42f ± 0.33        | 4.36d ± 0.33       | 4.21b ± 0.33   | 4.11c ± 0.33   | 4.23c ± 0.33  |
|                       | 1  | 6.56d ± 0.341       | 4.46d ± 0.34       | 4.23a ± 0.34   | 4.27bb ± 0.34  | 4.35bc ± 0.34 |

Note: Different capital letters; A, B, C, D, F indicate a statistical difference between the times at α = .05 level among products at each time. Different small letters; a, b, c, d, f indicate a statistical difference between the concentrations at α = .05 level among products at each time.

Abbreviation: WPH, whey protein hydrolysates.

Table 2: Effect of addition of *Spirulina platensis* and WPH added or not probiotics on the total solid and protein values after the storage time

| Addition | Total solid | Protein |
|----------|-------------|---------|
|          | *Spirulina* + WPH | *Spirulina* + WPH + Probiotic | *Spirulina* + WPH | *Spirulina* + WPH + Probiotic |
| 0        | 0.082       | 0.082   | 17.17   | 17.17   |
| 0.25     | 0.087       | 0.083   | 18.70   | 19.72   |
| 0.5      | 0.090       | 0.090   | 20.57   | 23.46   |
| 1        | 0.099       | 0.099   | 21.59   | 25.16   |

Note: Different capital letters; A, B, C, D, F indicate a statistical difference between the times at α = .05 level among products at each time. Different small letters; a, b, c, d, f indicate a statistical difference between the concentrations at α = .05 level among products at each time.

Abbreviation: WPH, whey protein hydrolysates.
due to the addition of protein-rich *S. platensis* biomass and WPH. It was reported that WPH contains about 45% to 50% of total milk solids.

### 3.7 The viscosity and Hunter color parameters

Effects of *S. platensis* + WPH added or not probiotics on the viscosity of ayran are shown in Table 3. Compared with the control samples, the addition of these supplements significantly decreased (*P* < .05) the viscosity value after the fermentation and storage time.

**Table 3** Effect of addition of *Spirulina platensis* and WPH added or not probiotics on the viscosity value through fermentation and storage time

| Sample                  | Concentration | After 1 day   | After 7 days   | After 14 days  | After 21 days  |
|-------------------------|---------------|---------------|---------------|---------------|---------------|
| Control                 | 0             | 42.24 ± 0.01  | 50.76 ± 0.01  | 57.81 ± 0.01  | 40.95 ± 0.01  |
| *Spirulina* + WPH       | 0.25          | 26.67 ± 0.01  | 36.57 ± 0.01  | 43.96 ± 0.01  | 40.32 ± 0.01  |
|                         | 0.5           | 27.23 ± 0.01  | 30.54 ± 0.01  | 36.04 ± 0.01  | 32.21 ± 0.01  |
|                         | 1             | 28.78 ± 0.01  | 29.76 ± 0.01  | 30.27 ± 0.01  | 24.33 ± 0.01  |
| *Spirulina* + WPH + Probiotic | 0.25       | 19.67 ± 0.01  | 21.78 ± 0.01  | 23.67 ± 0.01  | 22.62 ± 0.01  |
|                         | 0.5           | 20.05 ± 0.01  | 20.95 ± 0.01  | 21.87 ± 0.01  | 17.95 ± 0.01  |
|                         | 1             | 21.12 ± 0.01  | 21.56 ± 0.01  | 21.76 ± 0.01  | 18.78 ± 0.01  |

*Note: Different capital letters; A, B, C, D indicate a statistical difference between the times at α = .05 level among products at each time. Different small letters; a, b, c, d indicate a statistical difference between the concentrations at α = .05 level among products at each time.*

**Table 4** Effect of addition *Spirulina platensis* and WPH added or not probiotics on Hunter color parameters through fermentation and storage time

| Sample                  | %             | After 1 day   | After 7 days   | After 14 days  | After 21 days  |
|-------------------------|---------------|---------------|---------------|---------------|---------------|
| Lightness L* Control    | 0             | −2.58 ± 0.01  | −1.54 ± 0.01  | −1.24 ± 0.01  | −1.03 ± 0.01  |
| *Spirulina* + WPH       | 0.25          | −8.44 ± 0.01  | −8.37 ± 0.01  | −8.37 ± 0.01  | −7.83 ± 0.01  |
|                         | 0.5           | −9.87 ± 0.01  | −9.82 ± 0.01  | −9.82 ± 0.01  | −9.34 ± 0.01  |
|                         | 1             | −11.97 ± 0.01 | −11.55 ± 0.01 | −11.93 ± 0.01 | −11.51 ± 0.01 |
| *Spirulina* + WPH + Probiotic | 0.25       | −6.97 ± 0.01  | −7.72 ± 0.01  | −7.84 ± 0.01  | −7.47 ± 0.01  |
|                         | 0.5           | −8.92 ± 0.01  | −8.37 ± 0.01  | −8.88 ± 0.01  | −9.33 ± 0.01  |
|                         | 1             | −11.71 ± 0.01 | −11.74 ± 0.01 | −11.45 ± 0.01 | −11.43 ± 0.01 |
| Redness a* Control      | 0             | 3.95 ± 0.01   | 4.84 ± 0.01   | 5.82 ± 0.01   | 7.98 ± 0.01   |
| *Spirulina* + WPH       | 0.25          | 3.22 ± 0.01   | 3.17 ± 0.01   | 2.55 ± 0.01   | 3.54 ± 0.01   |
|                         | 0.5           | 2.97 ± 0.01   | 2.84 ± 0.01   | 2.47 ± 0.01   | 3.65 ± 0.01   |
|                         | 1             | 4.13 ± 0.01   | 4.16 ± 0.01   | 4.31 ± 0.01   | 4.08 ± 0.01   |
| *Spirulina* + WPH + Probiotic | 0.25       | 2.87 ± 0.01   | 2.83 ± 0.03   | 2.56 ± 0.01   | 3.23 ± 0.01   |
|                         | 0.5           | 2.73 ± 0.01   | 2.33 ± 0.01   | 2.65 ± 0.01   | 3.34 ± 0.01   |
|                         | 1             | 2.67 ± 0.01   | 2.93 ± 0.01   | 2.94 ± 0.01   | 2.65 ± 0.01   |

*Note: Different capital letters; A, B, C, D indicate statistical difference between the times at α = .05 level among products at each time. Different small letters; a, b, c, d indicate statistical difference between the concentrations at α = .05 level among products at each time.*

**Abbreviation:** WPH, whey protein hydrolysates.
fermentation and the storage times. The lowest the viscosity was found in the samples having \textit{S. platensis} + WPH with probiotics. The similar behavior in viscosity obtained from nonfat yogurt.\textsuperscript{30} However, Patocka et al\textsuperscript{31} recorded that the addition of WPH ranged from 1\% to 3\% did not affect the viscosity value up to 35 days of storage. Martelli et al\textsuperscript{11} found that the fermentation process improved the viscoelastic properties of skimmed milk and soy milk. In addition, they observed that viscosity of skimmed milk and soymilk decreased by the addition of \textit{S. platensis}. The effects of studied parameters on viscosity values could relate to different characteristics of physicochemical properties such as exopolysaccharides produced by the bacteria in the samples.

Hunter color parameters; \(L^*\) lightness-darkness, \(b^*\) yellowness-blueness, and \(a^*\) redness-greenness values have been reported to ascribe the gradation of the visual color.\textsuperscript{32} It can be declared that the color of the fermented product is a remarkable criterion for the preferences of consumers also for the shelf life. Hunter color values can give trustworthy knowledge about the quality and safety properties of foods.

As given in Table 4, the addition of \textit{S. platensis} + WPH added probiotics decreased \(L^*\) value (lightness) significantly \((P < .05)\) when it was compared with the control after the fermentation and at 14th and 21st days of the storage. Similar behavior was found in \(a^*\) (redness) and \(b^*\) (yellowness-blueness) levels (Table 4). The decrement in the \(L^*\) at these samples could be due to pigments such as c-phycocyanin and allophycocyanin in \textit{S. platensis} biomass.\textsuperscript{33} Martelli et al\textsuperscript{11} reported that the addition of \textit{S. platensis} showed a drastic impact on colorimetric characteristics of milk and soy fermented beverages.

4 | CONCLUSIONS

The addition of different levels of \textit{S. platensis} + WPH + probiotics into the ayran had the boosting effect on the growth of \textit{L. delbrueckii} subsp. \textit{bulgaricus}, \textit{B. lactis}, \textit{St. thermophilus}, and \textit{L. acidophilus}, and physico-chemical parameters of ayran throughout the fermentation period and the storage period. Variation in concentrations of these supplements cause to change the total solids and protein values in ayran and the highest levels were measured at the 1\% mixture of SP + WPH + probiotics. The encountered variability in the boosting effect of \textit{S. platensis} and WPH could depend on their addition values into ayran. These supplements had great potential for improving the growth of probiotic culture as well as the nutritional quality of ayran.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest regarding the publication of this article.

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