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Plant protein enrichment effect on the physical, chemical, microbiological, and sensory characteristics of yogurt

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Abstract

Yogurt is a coagulated dairy product that is fermented by typical starter cultures. Yogurt has the highest demand rate among fermented products due to its sensory properties. Well-consumed food products are being enriched by either adding nutrients or improving the already available content for the development of the product. From this point of view, protein enrichment is one of the approaches that contribute to the nutrient content of food products. The addition of plant-based protein is a step that improves the nutrient content and is a sustainable approach for the functionalization of the product. In the present study, two types of plant protein were added; chickpea and pea in two different concentrations of 0.5% (w/w) and 1% (w/w) to determine the changes in the physicochemical, textural, microbiological, and sensory properties of the yogurt during storage at 4°C for 21 days. In yogurt samples, pH value, titratable acidity, color measurements (L^*, a^*, b^*), texture parameters (firmness, consistency, cohesiveness), and microbiological contents show a significant difference during the storage period ($p < .05$). On the other hand, adding plant protein had no significant difference in syneresis value during storage ($p > .05$). The sensory evaluation results highlighted that protein enrichment develops consistency, mouthfeel, and oiliness scores compared to the control product (no plant protein added sample). These significant sensory modalities are determined by the casein network and water-binding capability of the proteins, which was visible as a result of this study.

Novelty impact statement:

- Plant protein addition improves the sensory properties of yogurt.
- Plant proteins are effective on the physicochemical properties of fermented dairy products.
- Functionalization of yogurt is achievable with the usage of plant protein addition.

1 | INTRODUCTION

Yogurt is defined as a coagulated dairy product fermented by typical starter cultures (Adolfsson et al., 2004). Among various dairy products, yogurt is one of the most popular and highly accepted in the global market (Kumar et al., 2015). It is not surprising to see yogurt as one of the leader products among the other foods due to its functional properties in terms of nutrition and product design and alterations. Functional foods are foods that include active compounds that have clinically proven health benefits (Martirosyan & Singh, 2015). These functional properties can derive from the natural content (of the bioactive ingredient), enrichment of the component, or removal of the unwanted compound. Yogurt is one of the naturally functional food which can also be boosted in terms of its functionality. Naturally, itself is known as a health promoter due to the starter culture content, which is also responsible for the appreciated sensory profile worldwide (Robinson et al., 2006). The fermentation and starter cultures and the nutritive compounds like; calcium, zinc, and vitamin B have a high impact on human health (El-Abbadi et al., 2014). Moreover, the natural functionality can be enriched with a few simple methods like fruit addition (do Espírito Santo et al., 2010; Sanchez-Segarra et al., 2000; Tarakçi & Kucukoner, 2003), probiotic enrichment (Cui et al., 2021; Molaee Parvarei et al., 2021), prebiotic additions (Balthazar et al., 2015; Esmaeilnejad Moghadam et al., 2019; Gustaw et al., 2011), fat reduction (Akgun et al., 2016; García-Gómez et al., 2019; Nikoofar et al., 2013), lactose reduction (Dekker et al., 2019; García-Gómez et al., 2019) and protein enrichment (Brückner-Gühmann et al., 2019; Keršienė et al., 2020).

Consumer demand for functional foods has accelerated since the beginning of the 2000s. Health enhancement, better nutrition, and higher nutrient control are the most important aims of consumers. Noteworthy, sustainable nutrition awareness forces producers and consumers to seek alternative sources of high-impact ingredients such as protein content. Following this approach, plant protein involvement in the food formulation could be an alternative source for the protein enrichment approach. Several researchers have focused on using plant sources such as lentil flour, soy flour (Zare et al., 2012), or rice (Kumari et al., 2015) for prebiotic fortification. Legumes should be considered a beneficial ingredient for their high nutrient value. The previous literature findings usually focus on the whole usage of the legumes. Whole legume addition will include high carbohydrate addition (Jukanti et al., 2012). However, isolation of plant proteins of the legumes could better impact the nutritional quality than the whole legume addition to the formulation. The addition of plant protein to the fermented dairy product formulation is expected to increase nutrient quality. However, it is also essential to test the physicochemical and sensory properties that might change. High protein yogurt still lacks a standard definition. Nevertheless, the Codex standard defines yogurt as the contention of a minimum of 2.7% milk protein (FAO, 2011). Meanwhile, after fermentation, concentrated fermented milk is a 5.6% protein content product (FAO, 2011). According to the literature, protein content and composition variations change important physical and sensory features due to gel network alterations (Mistry & Hassan, 1992; Schkoda et al., 2001; Tamime et al., 2014).

To our knowledge, there are no previous studies that have investigated the physicochemical, textural, and sensory properties of chickpea and pea protein addition to the yogurt. Besides its formulation impact, the sensory features are critical for the product development approach and need an increasing number of studies. Such fermented milk product provides the high nutritional value of proteins and other beneficial substances, while probiotics promote human gastrointestinal tract motility and benefit. These functional properties and sensory advantages of yogurt specifically aid to contribute to final properties that associate with consumer demand. Therefore, in the present study, the primary motivation was to highlight the increased plant protein content, which is expected to alter the physicochemical and sensory properties to some extent. Hence, in the present study, the addition of various concentrations of two different plant proteins to the yogurt product was investigated with the profiling of the physicochemical, microbiological, textural, and sensory properties.

2 | MATERIALS AND METHODS

2.1 | Materials

Raw cow milk was obtained from the local cattle farms at Iğdır (Turkey). Plant proteins were chickpea protein (FCPP-70C) and pea protein (FYPP-85B) which were purchased from AGT Food and Ingredients (Regina, SK, Canada). According to the manufacturer's specifications, the chickpea protein contained 74.7% (w/w) (dry basis) protein, 5.9% (w/w) moisture, 0.7% (w/w) crude fiber, 2.9% (w/w) ash, while pea protein contained 86.5% (w/w) (dry basis) protein, 7.1% (w/w) moisture, 0.8% (w/w) crude fiber, 3.3% (w/w) ash. The starter culture was YO Series (1:1 mixture of YO130 and YO248). Freeze-dried starter culture "Lactoferm YO Series" (YO-130 and YO-248) were supplied from Biochem (Italy). The composition of these thermophilic cultures is *Streptococcus salivarius* subsp. *thermophilus* and *Lactobacillus delbrueckii* subsp. *Bulgaricus*.

2.2 | Sample preparation

In the study, five groups of samples were prepared (Control [C], CPP0.5, CPP1, PP0.5, and PP1). Samples containing different concentrations of proteins were prepared separately. Protein-added milk samples were preheated to 40°C for plant protein dissolving purposes. The last part of the raw milk was the control sample and had no added plant protein. The samples with and without protein addition were heated to 90°C and held for 10 min for pasteurization. They were then cooled to 42°C for inoculation with 0.2 $g\,L^{-1}$ of starter culture (*Streptococcus salivarius* subsp. *thermophilus* and *Lactobacillus delbrueckii* subsp. *Bulgaricus*) and incubated at 42°C until pH reached 4.6. These samples were packed in 200 ml enclosed caps and stored for 21 days at 4°C. Yogurt samples were tested for their properties on the first, seventh, 14th, and 21st days. The samples were coded as illustrated in Table 1.

2.3 | pH, titratable acidity, and syneresis

According to Martin et al. (2010), pH values were measured with a multiparameter interface with a pH electrode SP10-R (Consort multiparameter analyzer C3040, Belgium). Meanwhile, percentage titratable acidity values of yogurt samples were measured with 0.1 N NaOH titration (IDF, 1982). The amount of syneresis was determined according to the method described by Lopes et al. (2019). According to this method, 10 g of sample was centrifuged with a centrifuge operating at 4°C (Shaking, HZQ-X300, China) for 10 min at 7870 g. The

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TABLE 1 The content and codes of plant protein enrichment yogurt samples

Sample code[a]	Chick pea protein (%)	Pea protein (%)
Control (C)	0	0
CPP0.5	0.5	0
CPP1	1	0
PP0.5	0	0.5
PP1	0	1

[a]C: Control, CPP0.5:0.5% (w/w) chickpea protein added sample, CPP1: 1% (w/w) chickpea protein added sample, PP0.5:0.5% (w/w) pea protein added sample, PP1: 1% (w/w) pea protein added sample.

clear supernatant was weighed, and the percentage of the syneresis was calculated from the ratio of expelled whey to yogurt mass.

2.4 | Instrumental color assessment

The color properties of the yogurt samples were measured with a Hunter-Lab color meter (Konica Minolta CR-140, Osaka, Japan) (Jrad et al., 2019). Color data were expressed in Hunter Lab Units L^*, a^*, and b^*, which indicate lightness, green (−) to red (+), and blue (−) to yellow (+) hue properties, respectively.

2.5 | Texture analyses

For the textural analysis of the yogurt samples, a method previously suggested by Buriti et al. (2014) was applied. For texture analysis, Stable Microsystem TA.XT2 analyzer (Surrey, UK) with a 5 kg load cell and an adapted penetration probe (cylindrical probe with 25 mm diameter) was used with a back extrusion test at 5 mm/s and 1 mm/s for initial speed force and test speed, respectively. Assessed modalities were firmness (g), consistency (g.s), and cohesiveness.

2.6 | Microbiological analyses

Lactic acid bacteria in yogurt samples were determined as described by (Bulut et al., 2021). *Streptococcus salivarius* subsp. *thermophilus* enumerations were made on M17 agar (Merck KGaA, Darmstadt, Germany) while, *Lactobacillus delbrueckii* subsp. *bulgaricus* enumerations were carried out on MRS agar (pH 6.5±0.2; Merck KGaA, Darmstadt, Germany). M17 plates were incubated at 37°C under conditions for 48 h. Meanwhile, MRS plates were incubated at 37°C under anaerobic conditions for 72 h with Anaerobic Jar (Merck 116387).

2.7 | Sensory analysis

Sensory tests involved informed consent collection and were carried out following the Code of Ethics of the World Medical Association (Declaration of Helsinki). Sensory analysis was carried out with plant protein added yogurt samples by a group of 10 panelists with a 15-point hedonic scale. Samples were presented with a three-digit code and a randomized, balanced block design in 200 ml containers and water for cleansing purposes. Panelists were asked to score; visual consistency, color, visual syneresis, fermentation odor, typical yogurt odor, firmness, sourness, sweetness, mouth-coating, consistency, chewability after taste, and general acceptability of the samples.

2.8 | Statistical analysis

All experiments were performed in duplicate using a complete factorial experimental design (the plant protein concentration and the storage period, 4x4). The data were analyzed using SPSS Statistical Software (version 20.0 for Windows, SPSS Inc., Chicago, Illinois). Duncan's multiple range test was conducted to detect differences between the treatment and storage period at a 95% significance level.

3 | RESULTS AND DISCUSSION

3.1 | Physicochemical analysis

During the storage, the physicochemical properties (pH, titratable acidity, syneresis, color) of the yogurts are shown in Table 2. The yogurt samples' pH and titratable acidity values varied from 3.88 to 4.58 and from 0.93% to 1.53%, respectively. The pH values decreased while the titratable acidity values increased during the storage time ($p<.05$). These findings agree with those who stated that shelf life decreases the pH and increases the titratable acidity (Glibowski et al., 2019; Zhang et al., 2019). Lactose fermentation during the shelf life produces dissociated lactic acid, which contributes to acidity level and decreases the pH value (Costa et al., 2015). Additionally, the buffering capacity of the caseins determines the pH fluctuation during the storage period (Martin et al., 2009). The effect of plant protein addition to yogurt did not cause significant changes in the pH values ($p>.05$).

Titratable acidity is significant for the sensory properties (Tamime & Deeth, 1980). As seen in Table 2, for the first day of the storage, the highest titratable acidity was obtained with 1% pea plant protein added sample (0.99%), while the lowest was 1% chickpea protein added sample (0.93%). For the seventh and 14th days of storage, titration values were higher than those of control, while there was no significant difference between the seventh and 21st days ($p>.05$). Within the end of the storage period, 0.5% concentrations of both plant proteins were higher titration values than the others. The overall increased pattern of the titratable acidity is due to slowing down the proteolytic activity of the yogurt bacteria due to added plant protein which increases the production of lactic acid (Bulut et al., 2021). A similar study to ours showed that titratable acidity increases in yogurt with the addition of factors that lower

TABLE 2 Effect of the plant protein addition level and storage time on some selected characteristics of the yogurt samples

	Storage time (days)	pH	Titratable acidity (%)	Syneresis (%)	Color properties		
					*L**	*a**	*b**
Control (C)[1]	1	4.37 ± 0.01^{dA}	0.94 ± 0.01^{bcB}	30.35 ± 2.05^{aA}	61.82 ± 0.01^{bA}	-3.34 ± 0.01^{eB}	8.31 ± 0.01^{bA}
	7	4.05 ± 0.01^{cB}	1.02 ± 0.04^{aB}	23.85 ± 1.34^{bB}	51.90 ± 0.42^{aB}	-2.50 ± 0.04^{cA}	5.75 ± 0.20^{abB}
	14	4.03 ± 0.02^{bB}	1.35 ± 0.01^{cA}	23.55 ± 0.07^{bB}	52.23 ± 0.64^{abB}	-2.35 ± 0.18^{aA}	5.62 ± 0.24^{abB}
	21	3.90 ± 0.05^{cC}	1.37 ± 0.04^{aA}	23.20 ± 0.42^{aB}	52.44 ± 0.43^{aB}	-2.40 ± 0.07^{cA}	5.54 ± 0.23^{aB}
CPP0.5	1	4.54 ± 0.09^{bA}	0.97 ± 0.03^{abB}	27.50 ± 2.69^{aA}	61.37 ± 0.04^{cA}	-3.10 ± 0.00^{cB}	7.83 ± 0.01^{dA}
	7	4.09 ± 0.01^{bB}	1.02 ± 0.02^{aB}	23.50 ± 0.14^{bA}	51.63 ± 0.08^{aB}	-2.26 ± 0.08^{bA}	5.10 ± 0.06^{cB}
	14	4.11 ± 0.01^{aB}	1.44 ± 0.1^{aA}	24.95 ± 1.06^{aA}	51.00 ± 1.08^{bB}	-2.34 ± 0.16^{aA}	5.17 ± 0.23^{bB}
	21	3.99 ± 0.01^{bC}	1.53 ± 0.09^{aA}	24.40 ± 0.57^{aA}	51.67 ± 0.40^{abB}	-2.33 ± 0.02^{bcA}	4.96 ± 0.20^{bB}
CPP1	1	4.44 ± 0.01^{cA}	0.93 ± 0.01^{cB}	26.70 ± 2.97^{aA}	61.13 ± 0.01^{eA}	-3.21 ± 0.00^{dB}	8.66 ± 0.02^{aA}
	7	4.02 ± 0.01^{dB}	1.06 ± 0.10^{aB}	26.30 ± 0.28^{aA}	51.80 ± 1.66^{aB}	-2.34 ± 0.16^{bcA}	6.06 ± 0.30^{aB}
	14	4.00 ± 0.01^{bB}	1.42 ± 0.04^{abA}	24.85 ± 0.35^{aA}	51.03 ± 0.07^{bB}	-2.33 ± 0.03^{aA}	5.73 ± 0.02^{aB}
	21	3.88 ± 0.02^{cC}	1.49 ± 0.06^{aA}	24.25 ± 1.77^{aA}	51.75 ± 0.25^{aB}	-2.43 ± 0.04^{cA}	5.77 ± 0.04^{aB}
PP0.5	1	4.58 ± 0.00^{aA}	0.95 ± 0.01^{bcD}	25.60 ± 0.28^{aA}	61.29 ± 0.01^{dA}	-2.94 ± 0.01^{bC}	7.70 ± 0.01^{eA}
	7	4.14 ± 0.02^{aB}	0.99 ± 0.01^{aC}	23.42 ± 0.02^{bB}	52.42 ± 0.37^{aBC}	-2.20 ± 0.03^{abA}	5.41 ± 0.06^{bcB}
	14	4.12 ± 0.01^{aB}	1.40 ± 0.01^{abB}	22.95 ± 0.07^{bC}	53.75 ± 1.42^{aB}	-2.41 ± 0.08^{aB}	5.51 ± 0.18^{abB}
	21	4.06 ± 0.01^{aC}	1.51 ± 0.01^{aA}	22.75 ± 0.07^{aC}	50.94 ± 0.08^{bcC}	-2.23 ± 0.01^{bA}	4.86 ± 0.12^{bC}
PP1	1	4.46 ± 0.01^{cA}	0.99 ± 0.01^{aC}	25.45 ± 0.03^{aA}	62.38 ± 0.01^{aA}	-2.62 ± 0.01^{aC}	7.91 ± 0.01^{cA}
	7	4.13 ± 0.01^{aB}	1.04 ± 0.01^{aC}	23.36 ± 0.06^{bB}	51.18 ± 0.10^{aC}	-1.98 ± 0.04^{aA}	5.08 ± 0.06^{cC}
	14	4.12 ± 0.02^{aB}	1.37 ± 0.01^{bcB}	22.66 ± 0.08^{bC}	51.61 ± 0.15^{abB}	-2.12 ± 0.02^{aB}	5.34 ± 0.11^{abB}
	21	4.02 ± 0.02^{abC}	1.48 ± 0.06^{aA}	22.50 ± 0.02^{aD}	50.84 ± 0.11^{cD}	-2.09 ± 0.04^{aB}	4.80 ± 0.04^{bD}

Notes: Values are expressed mean ± standard deviation; values followed by different letters (lower case letters indicate the same concentration-effect within the storage time) (capital letters indicate differences between different concentrations for the same storage time) for each column and factor are significantly different by Duncan's multiple range test ($p < .05$).

[1]C: Control, CPP0.5:0.5% (w/w) chickpea protein added sample, CPP1: 1% (w/w) chickpea protein added sample, PP0.5:0.5% (w/w) pea protein added sample, PP1: 1% (w/w) pea protein added sample.

proteolytic activity, such as mint and dill (Akbal, 2013; Alwazeer et al., 2020; Metry & Owayss, 2009). According to the standards and literature findings, titratable acidity should fit in a range of values. This range is determined between 0.80% to 1.60% in the TS 1330 (Turkish Standards Institute, 2006). Our highest findings, which were on the 21st day, fit the National Codex.

Syneresis can be defined as serum separation and is an important structural property of set-type yogurts (Olagunju et al., 2020). The serum separation is reported as a quality defect and is usually due to casein network rearrangements during the storage (Ramirez-Santiago et al., 2010; van Vliet et al., 1997). In order to increase the water-binding capacity of yogurt, the producers increase the dry matter or add a stabilizer into the milk (Vital et al., 2015). Since syneresis is a consumer complaintive quality issue, it must be controlled when possible (Purwandari et al., 2007; Salvador & Fiszman, 2004). On the other hand, it is accepted as one of the determinant quality factors for determining shelf-life and acceptance (Kiros et al., 2016; Sidira et al., 2017). The syneresis findings of the plant protein added yogurt samples are illustrated in Table 2. The syneresis value on the first day of storage was highest with the control sample (30.35%) while lowest for the PP1 sample (25.54%). According to these findings, there was no significant difference between the samples within the first day of storage and the control sample ($p > .05$). However, CPP0.5 and CPP1 samples had a significant difference for the seventh, 14th, and 21st days while PP0.5 and PP1 had differences following the 14th day ($p < .05$). Specifically, 21st-day values of syneresis were higher for CPP0.5 and CPP1 and, oppositely, lower for PP0.5 and PP1 samples. The hydrophobic bonds could be the triggering force for the higher syneresis value for the CPP samples. A similar finding was supported by research on grape extract usage for cheese production, which highlighted that hydrophobic bonds could be the responsible reason for this case (Han et al., 2011). Additionally, it is an expected scenario to observe a decrease in serum separation during storage due to metabolic activities of the starter culture and loss of net pressure of the protein matrix (Akın, 1998). Also, the removal of the pH value from the isoelectric point of casein and the interaction between proteins and water had an effect on the reduction of syneresis as well as the temperature of storage (Walstra & Jenness, 1984).

Color is an important quality factor for dairy products. Milk has a characteristic opaque white color which is mainly associated with casein proteins (Aryana et al., 2006). Colloid particles like fat globules and casein micelles have light scattering capability, which gives the characteristic white colors. Color properties of the yogurt samples are illustrated in Table 2 during the storage period. For the first day of production, the L^* value of the samples shows significantly different values ($p<.05$). The control sample had a higher L^* value throughout the storage time among the samples. At the end of the storage period, the L^* value decreased. PP0.5 and PP1 samples had an increasing trend on the 14th day compared to the seventh day of storage which was determined to be due to the green color pigments of the protein. This green pigment addition was reported to be contributing to the brightness (L^*) measure physically (Bulut et al., 2021). It is expected to see color changes throughout the storage period to some extent, mostly due to the heat processes that might release color products (García-Pérez et al., 2005). This is supported by a study done by Shokery et al. (2017), who used green tea and moringa extracts to enrich the set yogurt formulae, which showed that the extract addition leads to decreasing L^* and yellowish color on those products. Therefore, in agreement with that study, the present findings showed decreasing L^* value in the yogurt samples with plant protein addition, especially with pea protein. Table 2 involves the measurements of the a^* and b^* values of the samples. The a^* value had negative (greenness) values throughout the storage period had decreased. The loss of greenness is due to a decrease in the phenolic substances which turn into either colorless or brown color during the time (Karaaslan et al., 2011). With time, the color change has also been supported due to proteolysis based on the starter culture and probiotic activities (Costa et al., 2017). Concentration and type of the plant protein have shown a significant difference within the a^* value of the samples ($p<.05$). Similar to the L^* value findings, pea protein added samples had a significant difference compared to the other samples due to the green color of the protein extract. On the other hand, b^* values also had significant differences throughout the storage time ($p<.05$). The b^* values were all positive, while at the end of the storage period, CPP1 was the only sample that had a higher b^* value than the control sample.

3.2 | Texture analyses

Table 3 illustrates the textural attributes measured for the plant protein added yogurt samples for the first, seventh, 14th, and 21st days of the storage. According to this, firmness value ranges were between 115.91 g to 190.61 g for the control sample and PP1, respectively. The firmness values were significantly different throughout the storage period ($p<.05$). This finding is validated by the previous literature

TABLE 3 Textural properties of various concentrations of plant protein addition level during 21 days of storage on yogurt samples

	Storage time (days)	Firmness (g)	Consistency (g.s)	Cohesiveness
Control (C)[1]	1	86.97 ± 2.40^{bC}	2028.30 ± 6.55^{aB}	25.47 ± 0.71^{bB}
	7	95.61 ± 0.89^{cB}	2054.66 ± 22.62^{aAB}	29.29 ± 1.36^{aC}
	14	75.77 ± 2.09^{dD}	1861.06 ± 47.28^{aC}	21.54 ± 1.47^{aA}
	21	115.91 ± 2.89^{cA}	2112.66 ± 12.86^{cA}	28.51 ± 0.95^{aBC}
CPP0.5	1	104.74 ± 1.53^{aC}	1528.99 ± 39.99^{bC}	28.62 ± 1.24^{cA}
	7	112.77 ± 1.12^{bB}	1134.68 ± 0.97^{eD}	36.99 ± 0.88^{bC}
	14	126.16 ± 3.27^{aA}	1646.66 ± 14.51^{bB}	33.02 ± 0.33^{bB}
	21	103.00 ± 1.41^{eC}	1796.92 ± 6.41^{dA}	30.88 ± 0.27^{bB}
CPP1	1	104.86 ± 0.95^{aC}	1556.35 ± 6.53^{bB}	36.67 ± 1.75^{dA}
	7	119.35 ± 2.16^{aA}	1782.76 ± 1.67^{bA}	49.51 ± 1.17^{cC}
	14	113.66 ± 1.73^{bB}	1830.75 ± 1.29^{aA}	38.74 ± 1.68^{cAB}
	21	107.89 ± 1.50^{dC}	1825.76 ± 49.74^{dA}	40.92 ± 0.24^{cB}
PP0.5	1	74.74 ± 2.91^{cD}	1295.89 ± 6.94^{cC}	22.06 ± 1.14^{aA}
	7	92.68 ± 0.71^{cC}	1276.54 ± 0.71^{dD}	28.77 ± 0.71^{aB}
	14	103.29 ± 2.38^{cB}	1503.39 ± 3.69^{cB}	24.39 ± 2.12^{aA}
	21	161.51 ± 0.71^{bA}	2238.56 ± 0.71^{bA}	40.77 ± 0.71^{cC}
PP1	1	85.19 ± 2.81^{bD}	1541.73 ± 0.82^{bD}	19.28 ± 0.08^{aA}
	7	121.34 ± 1.21^{aC}	1651.37 ± 28.36^{cB}	29.81 ± 0.52^{aB}
	14	128.36 ± 2.55^{aB}	1599.36 ± 10.40^{bC}	30.11 ± 1.42^{bB}
	21	190.61 ± 0.83^{aA}	2771.96 ± 4.24^{aA}	44.21 ± 1.42^{dC}

Notes: Values are expressed mean ± standard deviation; values followed by different letters (lower case letters indicate the same concentration-effect within the storage time) (capital letters indicate differences between different concentrations for the same storage time) for each column and factor are significantly different by Duncan's multiple range test ($p<.05$).

[1]C: Control, CPP0.5:0.5% (w/w) chickpea protein added sample, CPP1: 1% (w/w) chickpea protein added sample, PP0.5:0.5% (w/w) pea protein added sample, PP1: 1% (w/w) pea protein added sample.

TABLE 4 *Streptococcus salivarius* subsp. *thermophilus* (ST) and *Lactobacillus delbrueckii* subsp. *bulgaricus* (LB) (log cfu/ml) counts of plant protein addition level during 21 days of storage

	Storage time (days)	ST (log cfu/ml)	LB (log cfu/ml)
Control (C)[1]	1	6.35 ± 0.14^{bcB}	6.23 ± 0.14^{cC}
	7	6.47 ± 0.18^{cB}	6.33 ± 0.11^{dC}
	14	6.68 ± 0.14^{cB}	7.78 ± 0.12^{aB}
	21	7.20 ± 0.03^{bA}	8.29 ± 0.13^{aA}
CPP0.5	1	6.80 ± 0.28^{abB}	7.61 ± 0.16^{aC}
	7	7.18 ± 0.03^{aAB}	7.70 ± 0.14^{bC}
	14	7.40 ± 0.17^{aA}	8.17 ± 0.21^{aB}
	21	7.00 ± 0.14^{bAB}	8.62 ± 0.03^{aA}
CPP1	1	6.14 ± 0.06^{cD}	6.83 ± 0.04^{bB}
	7	6.71 ± 0.01^{bC}	6.95 ± 0.10^{cAB}
	14	7.20 ± 0.07^{abB}	7.27 ± 0.23^{bA}
	21	7.80 ± 0.02^{aA}	7.31 ± 0.16^{bA}
PP0.5	1	7.15 ± 0.04^{aB}	7.65 ± 0.28^{aBC}
	7	7.37 ± 0.03^{aA}	8.40 ± 0.03^{aA}
	14	7.13 ± 0.04^{abB}	8.05 ± 0.07^{aAB}
	21	7.05 ± 0.06^{bB}	7.47 ± 0.18^{bC}
PP1	1	6.40 ± 0.28^{bcC}	6.24 ± 0.08^{cB}
	7	6.71 ± 0.02^{bBC}	6.96 ± 0.14^{cA}
	14	7.01 ± 0.01^{bAB}	7.20 ± 0.21^{bA}
	21	7.15 ± 0.07^{bA}	7.26 ± 0.11^{bA}

Notes: Values are expressed mean ± standard deviation; values followed by different letters (lower case letters indicate the same concentration-effect within the storage time) (capital letters indicate differences between different concentrations for the same storage time) for each column and factor are significantly different by Duncan's multiple range test ($p < .05$).

[1]C: Control, CPP0.5:0.5% (w/w) chickpea protein added sample, CPP1: 1% (w/w) chickpea protein added sample, PP0.5:0.5% (w/w) pea protein added sample, PP1: 1% (w/w) pea protein added sample.

findings which mention that the increasing shelf-life is a determinant factor for the increasing firmness (Vieira et al., 2019). Control, PP0.5, and PP1 samples showed a significant difference within the sample group during the shelf-life ($p < .05$). The thickener characteristic of the plant protein and extracts is an essential contributor to the final product's firmness behavior, and molecular interactions determine this property (Domagala et al., 2005). Specifically, on the 21st day of storage, whole samples have a significant difference ($p < .05$).

Consistency is the attribute related to the flowing characteristic of the sample. The consistent findings are illustrated in Table 3. The findings showed that the consistency value increased during storage time, especially with PP0.5 and CPP1 samples ($p < .05$). For the first day of storage, the control sample had the highest value among all, while on the 21st day, PP1 had the highest value. The addition of the plant protein shows a significant difference at the end of 21 days of shelf life compared to the control yogurt ($p < .05$).

The last textural property was cohesiveness which results are reported in Table 3. Cohesiveness can be defined as the resistance force between the food surface and the upper palate, tongue, and teeth (Fox et al., 2017). Cohesiveness value was found to be negative values, and with increasing storage, PP0.5 and PP1 samples had increasing cohesiveness ($p < .05$). Najgebauer-Lejko (2014) reported that increasing cohesiveness results from the protein–phenol interactions that strengthen the inner attractive forces, which increase cohesiveness. Except for the PP1 sample, cohesiveness values decreased after the 14th day of storage. This decrease could be related to lipolysis of the fats or disruption of the casein network of the dairy products with the storage (Delgado et al., 2011; Yates & Drake, 2007).

3.3 | Microbiological analyses

Streptococcus salivarius subsp. *thermophilus* (ST) and *Lactobacillus delbrueckii* subsp. *bulgaricus* (LB) (log cfu/ml) counts of the plant protein added to yogurt samples are presented in Table 4. According to these findings, the ST count of the whole samples was higher than those of the control until the end of the 14th day of storage ($p < .05$). One of the reasons for this may be due to the polyphenol content of plant proteins (Zhang et al., 2019). Control, CPP1, and PP1 samples had increasing numbers of ST throughout the storage period. At the end of the 21st day of storage, the CPP1 sample had significantly higher counts of ST ($p < .05$). Other samples, meanwhile, did not show a significant effect on the storage period compared to the control ($p > .05$).

LB counts of the plant protein added samples were higher than those of control at the first and seventh days of storage ($p < .05$). LB count is important for dairy products since it positively affects the digestibility and nutrition content (Shori, 2013). There was no significant difference between the samples at the end of the storage, except for the PP0.5 and control samples ($p > .05$). The highest LB count was found with the CPP0.5 sample at the end of the storage. Specifically, the lower protein content was found to show higher counts of LB comparing the two concentration ranges with each other. Considering the 21st day, LB counts, all samples containing protein plant showed lower numbers than those of control ($p < .05$). The numbers of ST and LB during the shelf life is vital since the consumer is seeking an active form of biota from the yogurt, which is determined by the pH, temperature, nutrients, and storage conditions (Deshwal et al., 2021; Michael et al., 2010; Shori, 2015).

3.4 | Sensory analyses

Average hedonic scores for color, syneresis, firmness, sourness, oiliness, consistency, mouthfeel, and general acceptance are shown in Table 5, and spider webs of the sensory modalities are plotted in a spider-web illustration in Figure 1. For the general acceptance, the first day of storage was the highest control sample. According to the results, 1 day of storage showed a significant difference for all tested attributes for the whole sample set ($p < .05$). Specifically, plant protein addition resulted in a bit drop in the scores of general

TABLE 5 Sensory evaluation of the plant protein added yogurt samples stored at 4°C for 21 days[1]

Samples	Storage time (days)	Color	Syneresis	Firmness	Sourness	Oiliness	Consistency	Mouthfeel	General acceptance
Control (C)[2]	1	4.8	5.1	5.2	4.2	5.6	5.7	8.9	8.8
	7	5.1	8.2	4.7	6.4	5.1	7.9	10	10.7
	14	5.4	6.6	6.2	5.8	6.8	7.6	9	7.6
	21	4.7	4.9	7.8	6.9	9.4	8.6	8.5	7.8
CPP0.5	1	5.2	7.2	4.7	5.4	5.5	6.3	9.6	7.4
	7	6.5	8.9	8.3	8.5	8.9	8.8	10.7	6.1
	14	5.6	8	7.4	7.6	6.2	8.2	10.8	4.2
	21	4.6	6.2	7.2	7.6	7.7	7.7	7.6	5.7
CPP1	1	6.9	7.2	5	4.5	7.4	6.5	9.3	7.8
	7	7	8.7	7.7	7.8	8.9	9.7	11.7	6.5
	14	6.4	8.8	6.6	8	6.6	8.6	11.6	4.8
	21	6.6	6.7	7.5	6.7	10.3	8.7	7.6	7.1
PP0.5	1	4.3	7.8	5.1	4.4	7.6	5.3	10	8.2
	7	5.2	8.5	7.8	7.2	9.1	9	11.6	8.4
	14	6.2	7.2	6.8	7.8	7.4	8.2	10.8	4.8
	21	5.9	5.6	7.9	7.2	9.1	8.1	7.2	7.4
PP1	1	5	8.2	5.6	4.5	8.8	6.4	10.8	7.6
	7	6	8.4	8.9	5.6	9.4	9.9	12.3	10.9
	14	6	7.4	8.2	6.6	8	9	12	5.4
	4,8	5.2	9	10.2	8.8	9.2	9.5	8.7	4.8

[1]Evaluation using a 9-point hedonic scale.

[2](C: Control, CPP0.5:0.5% (w/w) chickpea protein added sample, CPP1: 1% (w/w) chickpea protein added sample, PP0.5:0.5% (w/w) pea protein added sample, PP1: 1% (w/w) pea protein added sample).

liking. The effect of the storage, on the other hand, was another important criterion for the tested modalities. Generally, the scores dropped down significantly at the end of 21 days of storage ($p < .05$). This finding illustrates that by the end of shelf life, plant protein addition is likely to reduce the sensory scoring of the yogurt product, which could also be a determinant factor for product development. The effect of shelf life on sensory scoring was similar to our findings by previous researchers who tested other yogurt products such as; fruit-flavored yogurt (Tarakçi & Kucukoner, 2003), carrot yogurt (Salwa et al., 2004), and traditional yogurts (Alirezalu et al., 2019). At varying concentrations, chickpea protein and pea protein addition significantly altered the general sensory profile. The panelist's responses illustrated that the syneresis and appearance of properties are significantly different from those of the control throughout the shelf life. These findings of the modalities were more visible by the increased storage. However, the consistency, mouthfeel, and oiliness were rated to be better than the control product. These texture-related modalities were determined mainly by the casein network and the water-binding capability of the proteins.

Figure 1 illustrates the sensory scores for the protein plant added yogurt samples for their storage time from the first day to the 21st day. The context, as mentioned earlier, about the loss of sensory profile by the increasing storage is more visible considering the plots.

4 | CONCLUSION

Results obtained in the present study indicated that the plant protein isolates addition is technology-wise possible to enrich the nutrient quality and functionalization of the yogurt product. Our results showed that protein enrichment would also develop better consistency, mouthfeel, and oiliness scores compared to the control product. Hence, plant protein addition can be used to develop a sensory profile due to the casein network and water-binding capability of the proteins. Noteworthy, this research was done with the motivation of underlying the pinning principles of plant-sourced protein addition on the properties of fermented dairy products during the shelf life. Further investigations and product formulation trials are still necessary to have a complete image of functional dairy product development. Further investigations are essential for the final product formulation approvals.

AUTHOR CONTRIBUTIONS

Menekse Bulut: Conceptualization; data curation; formal analysis; methodology; writing – original draft; writing – review and editing. **Eda Adal:** Conceptualization; data curation; formal analysis; methodology; writing – original draft; writing – review and editing. **Tugba Aktar:** Conceptualization; data curation; formal analysis; methodology; writing – original draft; writing – review and editing.

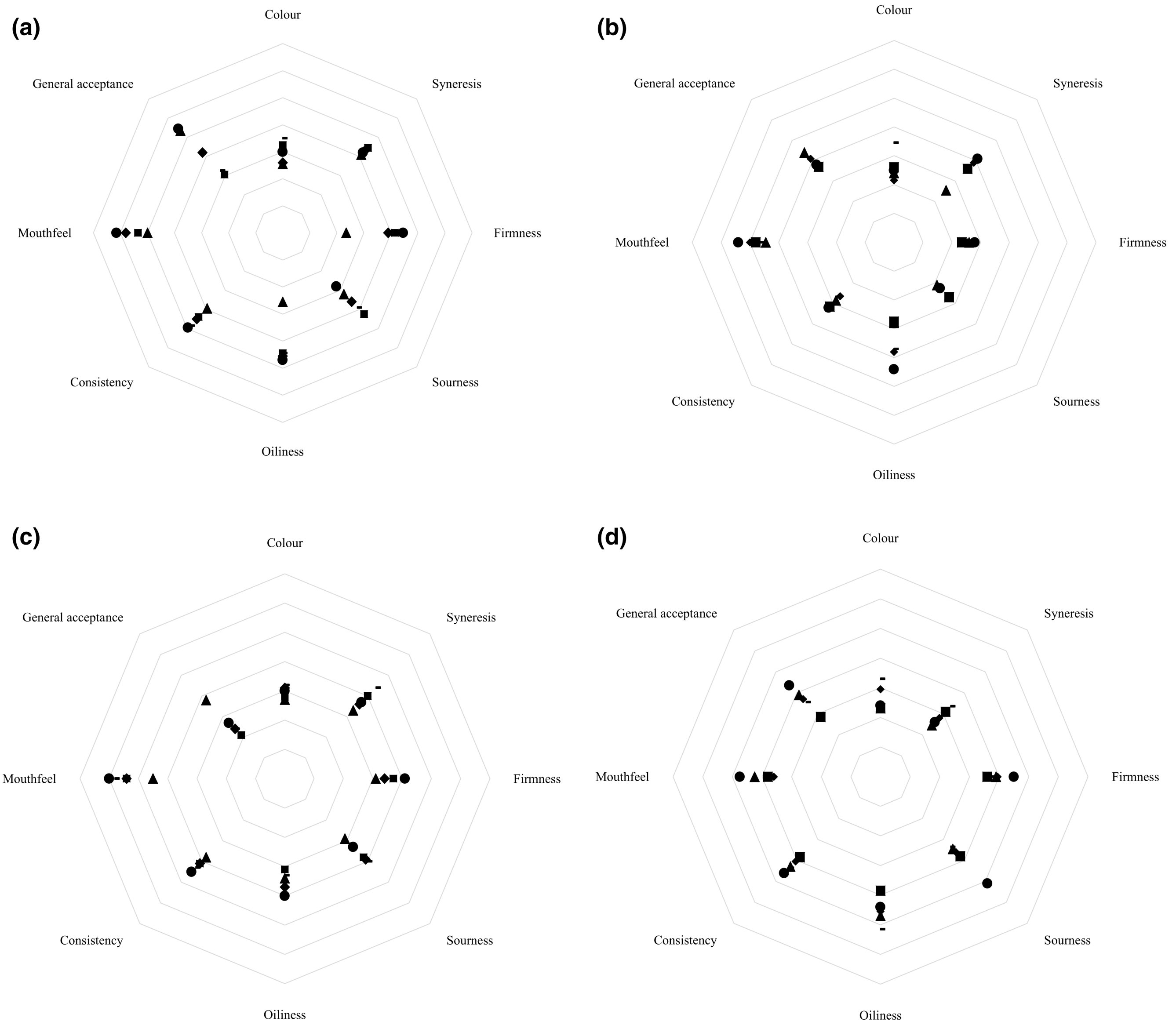

FIGURE 1 Sensory profiles of plant protein added yogurt samples stored at 4°C for 21 days. Individual attributes are positioned around a center (zero or not detected) point with the spokes representing attribute intensity scales, with higher (bigger score) values radiating outward. (a) Shows the sensory scoring on the first day of storage, (b) shows the sensory scoring on the seventh day of storage, (c) shows the sensory scoring on the 14th day of storage, and (d) shows the sensory scoring on the 21st day of storage. Sample codings are; C: Control sample (▲), CPP0.5:0.5% chickpea protein added sample (■), CPP1: 1% chickpea protein added sample (—), PP05:0.5% pea protein added sample (♦), PP1: 1% pea protein added sample (●).

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

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