

Article – Food /Feed Science and Technology

Effect of Flaxseed Powder on Physicochemical, Rheological, Microbiological and Sensory Properties of Yoghurt

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Editor-in-Chief: Alexandre Rasi Aoki

Associate Editor: Jéssica Caroline Bigaski Ribeiro

Received: 09-Jan-2021; Accepted: 28-Mar-2022.

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HIGHLIGHTS

- Effects of the addition of flaxseed powder (FSP) were examined on yoghurts.
- The FSP improved syneresis and water holding capacity of yoghurts.
- The FSP enhanced viscosity and rheological properties of yoghurts.
- Microbiological properties of FSP added yoghurts was coherent with legal.
- The sensory properties of FSP added yogurts were acceptable except for one containing 1%.

Abstract: In the presented research, five different ratios of flaxseed powder (FSP) (0, 0.25%, 0.50%, 0.75%, and 1%) were used in yoghurt production, and the samples were stored overall 28 days at refrigerator (+4 °C). The effect of FSP and the storage period on acidity, pH, syneresis, water holding capacity, rheological, viscosity, colorimetric, microbiological, and sensory properties of yoghurts were investigated. The acidity, syneresis values of yoghurt samples decreased with the addition of FSP, conversely, the pH, water holding capacity values of the samples increased. The increments in the viscosity (50 and 100 rpm) and consistency coefficient values were observed depending upon the FSP ratio except for the sample containing 1% FSP. All yoghurt samples had pseudoplastic flow behavior. It was determined that the colorimetric parameters of samples changed significantly with the addition of FSP. The L^* and white index values of samples containing FSP decreased. Other colorimetric parameters indicated significant differences based on the addition of FSP and storage. The addition of FSP was not caused any negative effect on counts of *Lactobacillus bulgaricus* and *Streptococcus thermophilus*. The *Lactobacillus bulgaricus*, *Streptococcus thermophilus* and moulds-yeasts counts were consistent with the Codex Alimentarius and the Turkish Food Codex. The sample containing 1% FSP had the lowest evaluation scores in terms of all sensory properties. Therefore, FSP should not use more than 0.75%. Using of the 0.25% FSP in yoghurts could recommend for improve physicochemical, rheological and sensory properties of yoghurts.

Keywords: flaxseed; consistency coefficient; microbiological properties; sensory properties; yoghurt.

INTRODUCTION

Yoghurt is a fermented dairy product by heat induced protein denaturation and lactic acid bacteria (*Streptococcus salivarius* subsp. *thermophilus* (*S. thermophilus*) and *Lactobacillus delbrueckii* subsp. *bulgaricus* (*L. bulgaricus*)). Yoghurt has a three-dimensional gel structure. Yoghurt, which has a standard texture, is difficult to produce [1].

The nutritional value, healthy and texture of yoghurt is important properties for desire of consumers [1], [2]. The stabilizers such as gelatin, carrageenan, pectin and starch have been used to improve texture of yoghurt and to reduce syneresis. These stabilizers lead to off flavor and texture, and they are not completely health [1]. Soft and consistency texture are desired in yoghurt by consumers [3]. The number of studies to improve texture properties of yoghurt have increased, recently [4]–[8]. But, using plant origin products in yoghurts can restrict growth of yoghurt bacteria due to their containing phenolic compounds and antibacterial components [6].

The flaxseed has a wide use field as oil, fibre source or food in industry. Flaxseed has gel production properties due to water-soluble polysaccharides. Thus, flaxseed has been used as a new ingredient in foods [9]. Additionally, flaxseed has beneficial effects on human health due to containing lignin, α -linolenic acid, water-soluble gel and diet fibre [10].

The flaxseed belongs to linum family, and its Latin name is *Linum usitatissimum*. The flaxseed contains 40-45% oil, 20-25% protein and 28% diet fibre. Flaxseed has protective effect against some diseases such as obesity, cardiovascular disorders, because of rich nutritional value [10]. The studies on products enriched with diet fibre, omega fatty acids, proteins, amino acids, vitamins, minerals have been increased interest in the products [11]. The flaxseed was used for yoghurt production at different type by researchers [8], [12]. Marand and coauthors [12] determined physicochemical, antioxidant, sensory properties, and fatty acid composition of yoghurts added flaxseed powder (FSP) (1, 3 and 5%) during 21 days. Mousavi and coauthors [8] investigated texture and sensory properties of yoghurts with flaxseed (2 and 4%) during 28 days. The researchers did not examine effects of flaxseed on yoghurt bacteria counts and rheological properties.

In this research, FSP was added to yoghurt at different ratios (0.25%, 0.50%, 0.75% and 1%), and the yoghurts were stored during 28 days. The objectives of the study were: (a) to investigate effects of FSP on pH and acidity values of yoghurt samples, (b) to assess rheological, syneresis, colorimetric, and sensory properties during storage, (c) to examine the viability of yoghurt bacteria in yoghurt added FSP throughout storage. All properties of yoghurt samples at 1st, 7th, 14th, 21st and 28th days of storage were investigated.

MATERIAL AND METHODS

Material

FSP (Natural Hekimce - Aksuvital Doğal Ürünler A.Ş.) was purchased from local markets in Gümüşhane, Turkey. Raw milk was obtained from Şiran Süt A.Ş. factory in Gümüşhane. Lyophilized strater culture (DVS YO-130) for using in yoghurt production was provided from Biochem s.r.l. (Rome, Italy).

Method

Raw milk was divided into 5 equal lots as 3 kg. Dry matter of milk was adjusted with non-fat milk powder as 15.5%. FSP was added to raw milk at different ratios such as 0.25%, 0.50%, 0.75%, 1%, and they were coded as FY2, FY5, FY7 and FY10, respectively. The yoghurt sample without FSP was coded as C. Then, raw milk was pasteurized (at 85 °C for 20 min), and chilled to 45 °C. The yoghurt culture was inoculated to samples in 0.02% (w/w) ratio and put into sterilized glass jars (200 mL). The incubation was carried out at 43±1 °C for 3 h, finished at pH 4.7. The samples were kept at refrigerator at 4 °C during 28 days. Analyzes were carried out at 1st, 7th, 14th, 21st and 28th days.

Acidity and pH

Acidity values of yoghurt samples were determined with titration method by using 0.1 N NaOH [13]. pH was measured with a pH meter (WTW 3110, Weilheim, Germany) [14].

Syneresis and water holding capacity

Twenty-five grams of samples were weighted and kept at +4 °C for 120 min to filter. The whey was measured as volume and explained as mL/25 g [15]. Water holding capacity (WHC) was measured by using a centrifuge (Nüve NF 200, Ankara). Samples were weighted as 10 g in falcon tubes [16]. The tubes were centrifuged at 2750 g at +4 °C for 30 min. The results were calculated according to following equation:

$$\text{WHC}(\%) = (1 - (\text{pellet weight}/\text{sample weight})) * 100 \quad (1)$$

Rheological properties and viscosity

Viscosity values of the yoghurt samples were determined with spindle no:5 at 50 and 100 rpm by a viscometer (Model DV-II Brookfield Engineering Laboratories, Stoughton, MA, USA). Power Law model (Ostwald-de-Waele) was used to derive rheological values. According to following equation, the flow behavior index, consistency coefficient and shear rate were derived from apparent viscosity (η), and were symbolized as n , K , $\dot{\gamma}$, respectively [17]:

$$\eta = K\dot{\gamma}^n \quad (2)$$

Colorimetric parameters

Color measurements of yoghurt samples were performed with a CR-200 Minolta colorimeter (Minolta Camera Co., Osaka, Japan). The L^* , a^* and b^* values were measured by colorimeter. Hue angle (H°) [18], the saturation (C^*) [19] and whiteness index (WI) values [20] were calculated using the L^* , a^* and b^* values.

Microbiological properties

Sterile 0.85% (w/v) NaCl solutions were used for decimal dilutions. The *L. bulgaricus* counts in the samples were determined using Man Ragosa Sharpe agar (MRS, Merck, Darmstadt, Germany). MRS agar was kept under anaerobic condition at 37 °C for 72 h, and colonies were enumerated. Anaerobic condition was performed using anaerocult sachet (AnaeroPack-Anaero, Mitsubishi Gas Chemical America, Inc., New York, NY). *S. thermophilus* colonies were determined on M17 agar (modified Rogosa; Merck, Darmstadt, Germany), after incubation at 37 °C for 48 h under aerobic conditions [21]. Acidified (10% tartaric acid) potato dextrose agar (PDA) was used to determine the yeasts and molds. PDA was incubated at 25 °C for 5 days [22].

Sensory properties

All yoghurt samples during storage were evaluated in terms of sensory features (appearance, body-texture, syneresis, flavor, mouthfeel, over acceptability) by 30 semi-trained panelists. The age of panelists is between 20 and 40, and 17 of them were women and 13 were men. Hedonic scale was used in the evaluation. Yoghurt samples were scored between 1 (strongly dislike) and 9 (extremely wonderful) by panelist at 1st, 7th, 14th, 21st and 28 days of storage [23]. This research was reviewed and confirmed by the Research Ethics Committee of Gümüşhane University (the confirmation no. 95674917-108.99-E.10275 and date 13/03/2020).

Statistical analysis

Statistical analysis was performed using SPSS 17 statistical software (SPSS Inc., Chicago, IL, USA). Effect of treatments and storage on yoghurt samples was analyzed by one-way variance analysis (ANOVA). Differences among the samples were determined by Duncan's multiple-range test.

RESULTS AND DISCUSSION

In the yoghurt production, raw cow's milk that had 12.6% dry matter, 3.2% fat, 0.91% ash, 0.17% acidity, 6.60 pH, and somatic cell count 461000 cell mL⁻¹ was used. Dry matter, ash and pH of FSP were determined as 90.47%, 4.64%, and 6.11, respectively

Acidity and pH of yoghurt samples added FSP

Whey separation in yoghurt is one of the textural and physical properties that have an important role in consumer acceptance and quality. The pH value is an important parameter on the textural and rheological properties of yoghurt [24], [25].

The FSP concentration and the storage had statistically significant effect ($P < 0.01$) on acidity and pH values of yoghurt samples. The lowest acidity values were detected in sample FY10 during storage except for 7th day (Table 1). Acidity values of all yoghurt samples indicated a decrease at the end of storage except for the samples FY7 and FY10. The acidity values of sample FY7 changed during storage, but the changes were not statistically significant ($P > 0.05$). pH values of all samples decreased on the 14th day of storage, then increased on the 21st day of storage (Table 1). The sample C had the lowest pH values during storage except for the 14 day of storage. Except from day 14, there is a trend to pH decrease in all samples, which is expected in yogurts due to post-acidification [24], [26]. The pH values of samples C, FY2, FY7 shown a decrease at the end of storage comparing to at the beginning of storage ($P < 0.05$). Marand and coauthors [12] determined fatty acid profile, physicochemical, antioxidant, and sensory characteristics of yoghurt samples added FSP at different ratios (1%, 3% and 5%). They found the lowest acidity and the highest pH values in control sample during storage. Göktepe and Akın [27] investigated antioxidant activity, physicochemical, textural, and sensory properties of set-type yoghurts added immature and whole wheat (1, 2 and 3%). They determined significantly changes in pH and acidity values depended upon the addition of immature and whole wheat, and storage period. These results were in line with ours. Similarly, Atik and coauthors [28] reported that pH and acidity values of yoghurt samples were affected by the addition of chia seed mucilage (1,2 and 3%) and storage.

Differences in acidity and pH values might have caused from increment of protein content. Because, changes in acidity and pH values of samples are occurred to balance of buffering effect of protein and reached desired acidity and pH values [12]. Yoghurt bacteria might have caused changes in acidity and pH values during storage [3], [26], [28]. Because, for growth of yoghurt bacteria, they metabolize some nutritional elements such as carbohydrate and protein, and they produce many products (lactic acid, acetic acid, fatty acids) [29], [30]. Additionally, it has been stated that the natural acid content of herbal products adding to yoghurt might be an effect on the acidity and pH values [3].

Table 1. Acidity and pH values of yoghurt samples (mean \pm SD)

Parameters	Storage	C	FY2	FY5	FY7	FY10
	time (days)					
Acidity (%)	1	0.93 \pm 0.01 ^{a,AB}	1.04 \pm 0.01 ^{a,BC}	1.10 \pm 0.03 ^{a,CD}	1.21 \pm 0.06 ^{a,D}	0.89 \pm 0.07 ^{ab,A}
	7	0.93 \pm 0.05 ^{a,A}	0.97 \pm 0.01 ^{a,B}	1.00 \pm 0.06 ^{a,B}	1.13 \pm 0.07 ^{a,C}	0.83 \pm 0.02 ^{a,A}
	14	1.13 \pm 0.11 ^{bc,B}	1.19 \pm 0.06 ^{b,B}	1.22 \pm 0.05 ^{b,B}	1.20 \pm 0.01 ^{a,B}	0.96 \pm 0.03 ^{b,A}
	21	1.21 \pm 0.02 ^{c,B}	1.23 \pm 0.04 ^{b,B}	1.21 \pm 0.02 ^{b,B}	1.27 \pm 0.08 ^{a,B}	0.92 \pm 0.06 ^{ab,A}
	28	1.01 \pm 0.02 ^{ab,B}	1.02 \pm 0.02 ^{a,B}	1.02 \pm 0.01 ^{a,B}	1.13 \pm 0.02 ^{a,C}	0.93 \pm 0.01 ^{ab,A}
pH	1	4.31 \pm 0.00 ^{c,A}	4.38 \pm 0.07 ^{b,AB}	4.37 \pm 0.03 ^{b,AB}	4.43 \pm 0.01 ^{c,B}	4.58 \pm 0.05 ^{c,C}
	7	4.25 \pm 0.01 ^{b,A}	4.31 \pm 0.01 ^{ab,AB}	4.33 \pm 0.02 ^{b,B}	4.35 \pm 0.03 ^{b,B}	4.52 \pm 0.03 ^{c,C}
	14	4.24 \pm 0.00 ^{a,A}	4.24 \pm 0.00 ^{a,A}	4.24 \pm 0.00 ^{a,A}	4.24 \pm 0.00 ^{a,A}	4.24 \pm 0.00 ^{a,A}
	21	4.25 \pm 0.00 ^{b,A}	4.29 \pm 0.01 ^{ab,B}	4.33 \pm 0.00 ^{b,C}	4.33 \pm 0.01 ^{b,C}	4.41 \pm 0.01 ^{b,D}
	28	4.26 \pm 0.01 ^{b,A}	4.28 \pm 0.01 ^{a,A}	4.35 \pm 0.02 ^{b,B}	4.36 \pm 0.02 ^{b,B}	4.51 \pm 0.04 ^{c,C}

^{a,b,c}: Different lower case letters in same column show significant differences ($p < 0.05$).

^{A,B,C}: Different upper case letters in same row show significant differences ($p < 0.05$). C: yoghurt without flaxseed powder, FY2: yoghurt containing 0.25% flaxseed powder, FY5: yoghurt containing 0.50% flaxseed powder, FY7: yoghurt containing 0.75% flaxseed powder, FY10: yoghurt containing 1% flaxseed powder.

Syneresis and WHC of yoghurt samples added FSP

The syneresis and WHC of the samples were affected by the addition of FSP and storage ($P < 0.01$). The syneresis and WHC of samples were presented in Table 2. Decreases in the syneresis and increases in WHC of samples were observed depending on FSP concentration. The control sample had higher syneresis and lower WHC compared to sample FY7 throughout the storage period. The syneresis values of samples FY2 and FY7 were not significantly change during storage (Table 2). The samples C and FY10 had higher syneresis values at the first day (7.25 and 4.90 mL 25 g⁻¹, respectively) comparing to at the 28th day of storage (5.40 and 1.63 mL 25 g⁻¹) ($P < 0.05$). The syneresis of control sample were higher than that of sample FY10 during storage except for on the 7th day of storage (Table 2). These results may have been caused by the high fiber and protein content of FSP. Because the high protein content might increase syneresis in yoghurt due to strong bond strength [31]. The obtained results were lower than those of reported by Erkaya-Kotan

[32] for yoghurt produced by the addition of orange fibre. Mousavi and coauthors [33]. reported that syneresis of stirred yoghurt increased with addition of flaxseed (2% and 4%).

Table 2. Syneresis and WHC values of yoghurt samples (mean±SD)

Parameters	Storage	C	FY2	FY5	FY7	FY10
	time (days)					
Syneresis (mL 25 g ⁻¹)	1	7.25±0.00 ^{b,B}	4.65±1.56 ^{a,AB}	3.50±0.35 ^{a,A}	3.50±0.00 ^{a,A}	4.90±1.56 ^{b,AB}
	7	5.40±0.57 ^{a,B}	3.15±0.92 ^{a,A}	2.85±0.35 ^{a,A}	2.55±0.49 ^{a,A}	5.48±0.39 ^{b,B}
	14	5.60±0.57 ^{a,B}	3.60±0.85 ^{a,A}	3.20±0.28 ^{a,A}	2.90±1.27 ^{a,A}	2.95±0.07 ^{a,A}
	21	5.60±0.85 ^{a,C}	5.40±0.28 ^{a,C}	5.30±0.14 ^{b,C}	3.50±0.71 ^{a,B}	2.05±0.07 ^{a,A}
	28	5.40±0.42 ^{a,C}	4.30±1.13 ^{a,BC}	3.10±0.28 ^{a,AB}	2.00±0.71 ^{a,A}	1.63±0.11 ^{a,A}
WHC (%)	1	28.90±6.93 ^{a,A}	41.90±0.99 ^{bc,AB}	46.35±12.52 ^{b,AB}	35.10±3.68 ^{c,AB}	50.75±3.89 ^{a,B}
	7	29.30±3.25 ^{a,AB}	35.60±0.28 ^{a,ABC}	43.80±5.66 ^{b,BC}	22.90±1.98 ^{ab,A}	54.25±14.50 ^{a,C}
	14	30.35±0.07 ^{a,AB}	47.60±0.14 ^{d,C}	32.15±0.21 ^{ab,B}	26.05±4.17 ^{b,A}	52.25±0.35 ^{a,C}
	21	29.15±0.07 ^{a,A}	43.00±0.71 ^{c,C}	42.75±1.06 ^{b,C}	36.70±0.71 ^{c,B}	50.50±0.71 ^{a,D}
	28	26.80±6.65 ^{a,A}	40.65±1.48 ^{b,A}	20.05±9.55 ^{a,A}	17.35±0.49 ^{a,B}	45.65±0.92 ^{a,B}

a,b,c: Different lower case letters in same column show significant differences ($p < 0.05$).

A,B,C: Different upper case letters in same row show significant differences ($p < 0.05$). C: yoghurt without flaxseed powder, FY2: yoghurt containing 0.25% flaxseed powder, FY5: yoghurt containing 0.50% flaxseed powder, FY7: yoghurt containing 0.75% flaxseed powder, FY10: yoghurt containing 1% flaxseed powder. WHC: Water holding capacity.

The centrifugation method is used to determine WHC, and it is different from syneresis during storage period. WHC indicates the mechanical stability of the protein matrix of yoghurt under G-force [2]. WHC of samples were between 28.90% and 50.75% at the first day, while that were between 26.80% and 45.65% at the end of storage. The sample FY10 had higher WHC than other samples on the 1st, 7th, 21st and 28th days of storage. The samples C and FY10 did not indicate significantly difference in terms of WHC during storage (Table 2). The WHC of samples FY2, FY5 and FY7 showed statistically significant decrements and increments during storage. Mousavi and coauthors [33]. and Marand and coauthors [12] reported that WHC improved in yoghurt samples added flaxseed. Öztürk and coauthors [34] found that WHC of yoghurt samples increased with the addition of oleaster flour (crusted and without crusted). The decrease of syneresis and increment of WHC in yoghurt samples added FSP might been resulted from increment of protein content and fibre amount. Because, water separation in yoghurt reduces due to protein interactions and fibre [12], [34].

Viscosity and rheological properties of yoghurt samples added FSP

Viscosity is an important parameter for the quality of yoghurt. It informs about properties of yoghurt such as firmness and consistency [33]. The FSP adding and the storage had significantly effect ($P < 0.05$) on viscosity (50 and 100 rpm) and consistency coefficient (K), while flow behavior index (n) values were only affected by storage period ($P < 0.05$). The lowest viscosity (50 and 100 rpm) values were determined in sample FY10 during storage. Viscosity values (50 and 100 rpm) of yoghurt samples indicated irregular changes during storage except for sample FY2 (at 50 rpm) and FY5 (at 100 rpm), but the changes were statistically significant ($P < 0.05$). The slightly increment in the viscosity values of samples based on FSP concentration until containing 1% was determined. FY5 and FY7 had usually high viscosity values and then decreased on FY10. These results might have been caused by strong gel strength depending on high FSP concentration. Marand and coauthors [12] and Mousavi and coauthors [33]. found that viscosity values of yoghurt samples added flaxseed were higher than control sample. These results are in line with our results. In addition, many factors such as the production process, used starter cultures, heat treatment, and formulation ratio could affect the viscosity values of yoghurt [12].

Table 3. Viscosity values and rheological properties of yoghurt samples (mean±SD)

Parameters	Storage time (days)	C	FY2	FY5	FY7	FY10
50 rpm	1	7148±204 ^{c,C}	7242±134 ^{a,C}	5780±258 ^{a,B}	7425±521 ^{ab,C}	3549±441 ^{d,A}
	7	6490±562 ^{bc,B}	6761±962 ^{a,B}	10212±1629 ^{b,C}	7971±152 ^{ab,BC}	2502±271 ^{bc,A}
	14	6166±251 ^{abc,B}	6043±734 ^{a,B}	9471±224 ^{b,C}	8582±811 ^{b,C}	1884±352 ^{ab,A}
	21	5149±589 ^{a,B}	6247±654 ^{a,BC}	8650±1410 ^{b,D}	7128±57 ^{a,CD}	1330±79 ^{a,A}
	28	5350±509 ^{ab,B}	5492±458 ^{a,B}	5240±1125 ^{a,B}	7318±359 ^{a,C}	3002±14 ^{c,d,A}
100 rpm	1	2730±226 ^{a,B}	2654±12 ^{a,B}	4321±184 ^{a,C}	4163±711 ^{ab,C}	782±80 ^{a,A}
	7	4449±293 ^{c,BC}	3740±271 ^{b,B}	5237±770 ^{a,C}	4170±225 ^{ab,BC}	1415±292 ^{bc,A}
	14	3665±413 ^{bc,B}	3102±53 ^{ab,B}	5080±192 ^{a,C}	5126±492 ^{b,C}	969±269 ^{ab,A}
	21	3197±427 ^{ab,B}	3593±121 ^{ab,B}	4671±606 ^{a,C}	4047±89 ^{a,BC}	609±20 ^{a,A}
	28	2525±34 ^{a,B}	4072±791 ^{b,C}	4132±25 ^{a,C}	4025±20 ^{a,C}	1522±109 ^{c,A}
<i>K</i> (Pa.s ⁿ)	1	4.00±0.52 ^{a,A}	4.05±0.49 ^{a,A}	4.25±0.29 ^{a,A}	4.31±0.29 ^{a,A}	3.38±0.57 ^{a,A}
	7	4.37±0.01 ^{a,B}	4.36±0.12 ^{a,B}	4.50±0.02 ^{a,B}	4.50±0.01 ^{a,B}	4.04±0.07 ^{ab,A}
	14	4.34±0.00 ^{a,B}	4.43±0.03 ^{a,BC}	4.52±0.00 ^{a,C}	4.53±0.02 ^{a,C}	3.88±0.11 ^{ab,A}
	21	4.27±0.03 ^{a,B}	4.37±0.10 ^{a,BC}	4.51±0.00 ^{a,D}	4.50±0.01 ^{a,CD}	3.78±0.04 ^{ab,A}
	28	4.35±0.14 ^{a,B}	4.41±0.04 ^{a,B}	4.44±0.04 ^{a,B}	4.49±0.04 ^{a,B}	4.10±0.03 ^{b,A}
<i>n</i>	1	0.40±0.11 ^{a,A}	0.34±0.01 ^{b,A}	0.33±0.05 ^{b,A}	0.35±0.13 ^{a,A}	0.24±0.01 ^{a,A}
	7	0.27±0.03 ^{a,A}	0.26±0.02 ^{a,A}	0.25±0.02 ^{a,A}	0.28±0.00 ^{a,AB}	0.33±0.03 ^{b,B}
	14	0.27±0.02 ^{a,A}	0.29±0.03 ^{ab,AB}	0.26±0.01 ^{ab,A}	0.27±0.00 ^{a,A}	0.33±0.01 ^{b,B}
	21	0.26±0.02 ^{a,A}	0.27±0.02 ^{a,A}	0.27±0.02 ^{ab,A}	0.29±0.00 ^{a,A}	0.36±0.00 ^{b,B}
	28	0.31±0.04 ^{a,A}	0.29±0.02 ^{ab,A}	0.29±0.02 ^{ab,A}	0.29±0.01 ^{a,A}	0.33±0.02 ^{b,A}
<i>r</i> ²	1	0.984±0.008	0.995±0.001	0.993±0.003	0.979±0.008	0.995±0.002
	7	0.987±0.011	0.983±0.005	0.986±0.007	0.975±0.007	0.987±0.007
	14	0.993±0.009	0.982±0.009	0.977±0.012	0.982±0.006	0.978±0.011
	21	0.997±0.006	0.985±0.008	0.978±0.009	0.978±0.007	0.978±0.009
	28	0.964±0.004	0.985±0.005	0.992±0.002	0.965±0.008	0.970±0.006

a,b,c: Different lower case letters in same column show significant differences ($p < 0.05$).

A,B,C: Different upper case letters in same row show significant differences ($p < 0.05$). C: yoghurt without flaxseed powder, FY2: yoghurt containing 0.25% flaxseed powder, FY5: yoghurt containing 0.50% flaxseed powder, FY7: yoghurt containing 0.75% flaxseed powder, FY10: yoghurt containing 1% flaxseed powder. *K*: Consistency coefficient, *n*: Flow behaviour index

Yogurt shows non-Newtonian flow (pseudoplastic). This behavior is affected by some factors such as shear-thinning, yield stress, viscoelasticity, and time-dependency. The rheological properties could be calculated using Brookfield viscosity [2]. *K* values of the samples did not change on the first day of storage ($P > 0.05$). *K* value of sample FY10 was lower than other samples during storage except for on the 1st day of storage (Table 3). *K* values of yoghurt samples were not significantly ($P > 0.05$) difference during storage except for sample FY10. *n* value of sample FY10 was higher than other samples containing FSP on the 7th, 14th and 21st days of storage. All yoghurt samples indicated pseudoplastic behavior ($0 < n < 1$). It was stated that WHC and viscosity values of yoghurt samples with dietetic fibre increased [12], [33]. In this research, significant differences in viscosity and rheological properties may be caused by the high fibre content of flaxseed [35], [36]. The dry matter content, starter culture, thickeners have important effect on the viscoelasticity properties of yoghurt [2]

Color parameters of yoghurt samples added FSP

The addition of FSP and the storage had significant effect on the *L*^{*} ($P < 0.01$; $P < 0.01$), *a*^{*} ($P < 0.01$; $P < 0.05$), *b*^{*} ($P < 0.01$; $P < 0.01$), *C*^{*} ($P < 0.01$; $P < 0.01$) and WI ($P < 0.01$; $P < 0.01$) values of yoghurt samples. *H*^o values were affected by only storage period ($P < 0.01$). *L*^{*}, *b*^{*}, *C*^{*}, *H*^o and WI values of yoghurt samples decreased with FSP rates. Conversely, *a*^{*} values of samples increased. *L*^{*} and WI values of samples C, FY5 and FY10 at the beginning of storage were higher than at the end of storage ($P < 0.05$). In general, the control sample had the highest *L*^{*}, *b*^{*}, *C*^{*}, *H*^o, and WI values during the storage period, had the lowest *a*^{*} values. It

was not determined significantly differences at H° value of all samples during the storage ($P>0.05$) except for sample FY7. Differences in color measurement values of the sample FY2 were no statistically significant during storage except for b^* and C^* values. The similar results were detected in previously some researches [12], [37–39]. It can be explained that many factors such as reduce of water content with fibre, destabilization of casein with pasteurization, changes in pH values and color pigments of plants, have effect color parameters of yoghurt [40]. In this study, pH values of samples increased (Table 1), while L^* and WI values decreased (Table 4). Additionally, a^* and b^* values changed significantly in samples added FSP based on color pigments content of FSP.

Table 4. Colorimetric parameters of yoghurt samples (mean \pm SD)

Parameters	Storage time (days)	C	FY2	FY5	FY7	FY10
L^*	1	98.98 \pm 0.64 ^{b,B}	96.69 \pm 1.91 ^{a,AB}	94.83 \pm 1.56 ^{c,AB}	91.85 \pm 3.56 ^{c,A}	91.73 \pm 2.86 ^{b,A}
	7	89.20 \pm 4.96 ^{a,A}	87.65 \pm 7.15 ^{a,A}	85.14 \pm 4.65 ^{a,A}	81.45 \pm 3.04 ^{a,A}	89.06 \pm 0.53 ^{ab,A}
	14	95.98 \pm 0.42 ^{ab,D}	90.48 \pm 0.06 ^{a,BC}	91.96 \pm 0.06 ^{bc,C}	89.56 \pm 1.30 ^{bc,B}	85.88 \pm 0.88 ^{a,A}
	21	95.89 \pm 0.91 ^{ab,D}	92.64 \pm 0.04 ^{a,C}	86.91 \pm 0.31 ^{ab,B}	85.06 \pm 0.88 ^{ab,A}	84.40 \pm 0.12 ^{a,A}
	28	94.41 \pm 3.16 ^{ab,B}	90.30 \pm 3.07 ^{a,AB}	88.33 \pm 1.49 ^{ab,AB}	87.52 \pm 1.79 ^{abc,AB}	83.87 \pm 3.11 ^{a,A}
a^*	1	-4.94 \pm 0.21 ^{a,A}	-2.94 \pm 0.59 ^{a,B}	-1.64 \pm 0.15 ^{a,BC}	-1.13 \pm 1.10 ^{a,C}	-0.93 \pm 0.10 ^{a,C}
	7	-4.97 \pm 0.23 ^{a,A}	-3.52 \pm 1.82 ^{a,AB}	-1.49 \pm 0.47 ^{a,B}	-1.03 \pm 0.86 ^{a,B}	-1.65 \pm 0.27 ^{a,B}
	14	-4.40 \pm 0.24 ^{b,A}	-2.12 \pm 0.37 ^{a,B}	-1.10 \pm 0.58 ^{a,BC}	-0.62 \pm 0.40 ^{a,C}	-1.44 \pm 0.38 ^{a,BC}
	21	-4.17 \pm 0.07 ^{b,A}	-2.81 \pm 0.08 ^{a,B}	-0.03 \pm 0.14 ^{b,D}	0.08 \pm 0.12 ^{a,D}	-1.53 \pm 0.88 ^{a,C}
	28	-4.40 \pm 0.04 ^{b,A}	-2.13 \pm 0.19 ^{a,B}	-1.10 \pm 0.42 ^{a,C}	-0.94 \pm 0.04 ^{a,C}	-1.00 \pm 0.01 ^{a,C}
b^*	1	13.96 \pm 0.36 ^{ab,C}	12.35 \pm 0.54 ^{a,B}	11.28 \pm 0.72 ^{b,AB}	10.75 \pm 0.35 ^{a,A}	11.48 \pm 0.60 ^{b,AB}
	7	16.12 \pm 2.41 ^{b,B}	14.28 \pm 0.18 ^{b,AB}	12.60 \pm 0.30 ^{c,A}	11.16 \pm 0.98 ^{a,A}	12.66 \pm 0.24 ^{c,A}
	14	13.70 \pm 0.48 ^{ab,B}	11.47 \pm 0.62 ^{a,A}	11.22 \pm 0.16 ^{b,A}	11.15 \pm 1.03 ^{a,A}	10.53 \pm 0.48 ^{a,A}
	21	8.93 \pm 3.27 ^{a,A}	11.36 \pm 0.12 ^{a,A}	9.25 \pm 0.17 ^{a,A}	9.66 \pm 0.10 ^{a,A}	9.93 \pm 0.06 ^{a,A}
	28	14.08 \pm 1.17 ^{b,B}	11.66 \pm 0.11 ^{a,A}	11.03 \pm 0.39 ^{b,A}	10.59 \pm 0.15 ^{a,A}	11.98 \pm 0.01 ^{bc,A}
H°	1	109.50 \pm 1.19 ^{a,C}	103.36 \pm 2.05 ^{a,BC}	98.24 \pm 0.22 ^{a,AB}	96.50 \pm 5.39 ^{a,AB}	94.62 \pm 0.26 ^{a,A}
	7	110.23 \pm 0.61 ^{a,C}	103.55 \pm 0.97 ^{a,BC}	91.70 \pm 2.23 ^{a,A}	92.90 \pm 3.14 ^{a,A}	98.51 \pm 5.08 ^{a,AB}
	14	107.81 \pm 0.32 ^{a,C}	100.52 \pm 2.35 ^{a,B}	95.54 \pm 2.81 ^{a,AB}	93.16 \pm 1.87 ^{a,A}	97.76 \pm 1.67 ^{a,AB}
	21	121.80 \pm 15.88 ^{a,AB}	102.41 \pm 0.51 ^{a,AB}	135.20 \pm 62.76 ^{a,AB}	179.59 \pm 0.63 ^{b,B}	98.71 \pm 4.89 ^{a,A}
	28	107.60 \pm 1.80 ^{a,C}	98.24 \pm 0.80 ^{a,B}	95.73 \pm 2.36 ^{a,A}	95.05 \pm 0.30 ^{a,A}	94.77 \pm 0.04 ^{a,A}
C^*	1	14.61 \pm 0.02 ^{b,C}	12.70 \pm 0.66 ^{b,B}	11.40 \pm 0.73 ^{a,AB}	10.84 \pm 0.24 ^{a,A}	11.52 \pm 0.60 ^{ab,AB}
	7	12.06 \pm 0.10 ^{ab,A}	11.14 \pm 0.13 ^{a,A}	10.38 \pm 1.65 ^{a,A}	9.88 \pm 0.92 ^{a,A}	11.06 \pm 1.20 ^{ab,A}
	14	14.39 \pm 0.53 ^{b,B}	11.67 \pm 0.54 ^{ab,A}	11.28 \pm 0.22 ^{a,A}	11.17 \pm 1.05 ^{a,A}	10.63 \pm 0.53 ^{ab,A}
	21	10.17 \pm 2.60 ^{a,A}	11.42 \pm 0.25 ^{a,A}	9.25 \pm 0.17 ^{a,A}	9.67 \pm 0.11 ^{a,A}	10.07 \pm 0.19 ^{a,A}
	28	14.75 \pm 1.10 ^{b,B}	11.86 \pm 0.13 ^{ab,A}	11.09 \pm 0.35 ^{a,A}	10.63 \pm 0.14 ^{a,A}	12.02 \pm 0.01 ^{b,A}
WI	1	85.11 \pm 0.21 ^{ab,A}	86.79 \pm 0.16 ^{a,A}	87.42 \pm 0.01 ^{c,A}	86.15 \pm 2.49 ^{a,A}	85.39 \pm 0.75 ^{b,A}
	7	87.23 \pm 0.22 ^{ab,D}	86.12 \pm 0.28 ^{a,CD}	82.91 \pm 1.93 ^{a,AB}	83.58 \pm 1.21 ^{a,BC}	80.09 \pm 1.31 ^{a,A}
	14	85.05 \pm 0.39 ^{ab,B}	84.93 \pm 0.39 ^{a,B}	86.14 \pm 0.13 ^{bc,B}	84.70 \pm 1.66 ^{a,B}	82.31 \pm 0.38 ^{ab,A}
	21	88.96 \pm 2.65 ^{b,C}	86.16 \pm 0.13 ^{a,BC}	83.97 \pm 0.35 ^{ab,AB}	82.20 \pm 0.67 ^{a,A}	81.42 \pm 0.01 ^{a,A}
	28	84.10 \pm 2.13 ^{a,A}	84.51 \pm 1.85 ^{a,A}	83.88 \pm 1.34 ^{ab,A}	83.59 \pm 1.46 ^{a,A}	79.84 \pm 2.49 ^{a,A}

a,b,c: Different lower case letters in same column show significant differences ($p<0.05$).

A,B,C: Different upper case letters in same row show significant differences ($p<0.05$). C: yoghurt without flaxseed powder, FY2: yoghurt containing 0.25% flaxseed powder, FY5: yoghurt containing 0.50% flaxseed powder, FY7: yoghurt containing 0.75% flaxseed powder, FY10: yoghurt containing 1% flaxseed powder. WI: white index.

Microbiological properties of FSP added yoghurt samples

The microbiological properties of samples are presented in Table 5. The storage time had significantly effect ($P<0.01$) on *L. bulgaricus* and *S. thermophilus* counts of samples. There were insignificant differences among yoghurt samples throughout the storage period ($P>0.05$). The *L. bulgaricus* counts of yoghurt samples were between 6.96 and 8.43 log CFU g⁻¹ (Table 5), but the differences were not significant ($P>0.05$). The *L. bulgaricus* counts of all samples significantly ($P<0.05$) decreased at the end of storage except for control

sample. The samples FY2 and FY5 had the highest *L. bulgaricus* counts on the 7th day of storage. *L. bulgaricus* counts of samples FY2, FY7 and FY10 decreased ($P < 0.05$) on the 28th day of storage. Mihoubi and coauthors [41] found that *L. bulgaricus* count of the yoghurt samples containing %3 FSP was slightly higher than control samples. Our results were not in agreement with Mihoubi and coauthors [41]. Erkaya-Kotan [32] reported that *L. bulgaricus* counts of yoghurt samples containing orange fibre were between 5.76 and 6.63 log CFU g⁻¹ during 21-day storage. The results by Erkaya-Kotan [32] were lower than those of our results.

Table 5. Microbiological properties of yoghurt samples

Parameters	Storage time (days)	C	FY2	FY5	FY7	FY10
<i>L. bulgaricus</i> (log CFU g ⁻¹)	1	8.22±0.65 ^{a,A}	8.21±0.26 ^{bc,A}	8.38±0.20 ^{bc,A}	8.24±0.17 ^{b,A}	8.40±0.00 ^{b,A}
	7	8.43±0.00 ^{a,A}	8.36±0.10 ^{c,A}	8.64±0.04 ^{c,A}	8.52±0.00 ^{b,A}	8.26±0.31 ^{b,A}
	14	6.94±1.12 ^{a,A}	7.46±0.89 ^{a,A}	8.41±0.28 ^{bc,A}	8.44±0.03 ^{b,A}	7.55±0.50 ^{ab,A}
	21	6.96±1.36 ^{a,A}	7.80±0.03 ^{bc,A}	7.89±0.03 ^{ab,A}	7.88±0.23 ^{ab,A}	8.01±0.45 ^{ab,A}
	28	7.37±0.07 ^{a,A}	7.70±0.44 ^{ab,A}	7.47±0.44 ^{a,A}	7.46±0.46 ^{a,A}	7.27±0.01 ^{a,A}
<i>S. thermophilus</i> (log CFU g ⁻¹)	1	8.40±0.45 ^{a,A}	8.25±0.48 ^{ab,A}	8.44±0.33 ^{a,A}	8.54±0.20 ^{a,A}	8.35±0.02 ^{ab,A}
	7	8.67±0.06 ^{a,B}	8.68±0.05 ^{bc,B}	8.68±0.04 ^{a,B}	8.69±0.01 ^{a,B}	8.19±0.10 ^{ab,A}
	14	7.16±1.65 ^{a,A}	8.26±0.15 ^{ab,A}	7.15±1.63 ^{a,A}	8.56±0.08 ^{a,A}	7.46±1.02 ^{a,A}
	21	7.47±1.25 ^{a,A}	8.16±0.22 ^{a,A}	8.36±0.02 ^{a,A}	8.41±0.00 ^{a,A}	8.47±0.01 ^{ab,A}
	28	9.05±0.02 ^{a,A}	8.92±0.16 ^{c,A}	9.07±0.07 ^{a,A}	9.05±0.10 ^{b,A}	9.13±0.02 ^{b,A}

a,b,c: Different lower case letters in same column show significant differences ($p < 0.05$).

A,B,C: Different upper case letters in same row show significant differences ($p < 0.05$). C: yoghurt without flaxseed powder, FY2: yoghurt containing 0.25% flaxseed powder, FY5: yoghurt containing 0.50% flaxseed powder, FY7: yoghurt containing 0.75% flaxseed powder, FY10: yoghurt containing 1% flaxseed powder.

Significantly differences in *S. thermophilus* counts were not determined ($P > 0.05$) during storage except for the 7th day of storage (Table 5). The addition of FSP did not lead to a negative effect on *S. thermophilus* counts of samples. The differences in *S. thermophilus* counts of samples C and FY5 were no significant ($P > 0.05$) throughout the storage period. *S. thermophilus* counts of samples FY2, FY7 and FY10 were the highest at the end of storage. *L. bulgaricus* and *S. thermophilus* counts of the sample C did not indicate significantly change during storage. Demirkol and Tarakci [42] determined that the addition of dried grape (*Vitis labrusca* L.) pomace did not have significant effect on *L. bulgaricus* and *S. thermophilus* counts of yoghurt samples. These results were in line with those of the presented research. Conversely, some researchers found that *L. bulgaricus* and *S. thermophilus* counts of yoghurt samples containing green tea and Jerusalem artichoke powder increased compared to control samples [43], [44]. Bioactive compounds such as phenolic components could cause differences in starter culture activity of yoghurt [42], [45]. So, bioactive compounds of the ingredient might have affected the starter culture activity of yoghurt samples.

L. bulgaricus and *S. thermophilus* counts of yoghurt samples must be at least 10⁷ CFU g⁻¹ according to Turkish Food Codex [46] and the Codex Alimentarius [47]. *L. bulgaricus* and *S. thermophilus* counts of all yoghurt samples were in accordance with the Turkish Food Codex [46] and the Codex Alimentarius [47].

Molds-yeasts counts of all yoghurt samples were determined as <2 log CFU g⁻¹ overall 28 days of storage. These results comply with reported by Göktepe and Akın [27]. The molds-yeasts counts are an indicator of contamination and are one of the most important problems in fruity yoghurts [48]. According to Turkish Food Codex [46] and Codex Alimentarius [47], yoghurts must not have the molds-yeasts. All yoghurt samples were coherent with codex [46], [47]. It was reported that phenolic compounds could be inhibited the growth of molds-yeasts [48]. The flaxseed had rich in terms of bioactive compounds such as phenolics [49].

Sensory properties of yoghurt samples added FSP

Sensory scores were showed in Figure 1. The FSP had a significant effect ($P < 0.01$) on appearance, body and texture, flavor, mouth feel and overall acceptability scores, meanwhile storage period ($P < 0.01$) had a significant effect on appearance and body and texture scores. In general, samples C and FY2 were more appreciated than other samples in terms of all sensory properties during storage. The sample containing 1% FSP were the least desirable sample. The decrease in all sensory scores of samples was observed depending on FSP concentration. Sensory scores of yoghurt samples decreased at the end of storage

compared to at the beginning of storage. Similar results were reported by Marand and coauthors [12] and Mousavi and coauthors [8]. It was reported that phenolic components had effects on sensory properties such as bitter and pungent taste in foods [50]. In this research, sensory properties might have affected by rich phenolic components content of flaxseed, and the decrement in sensory scores at the end storage could have caused from more dissolved of phenolic compounds.

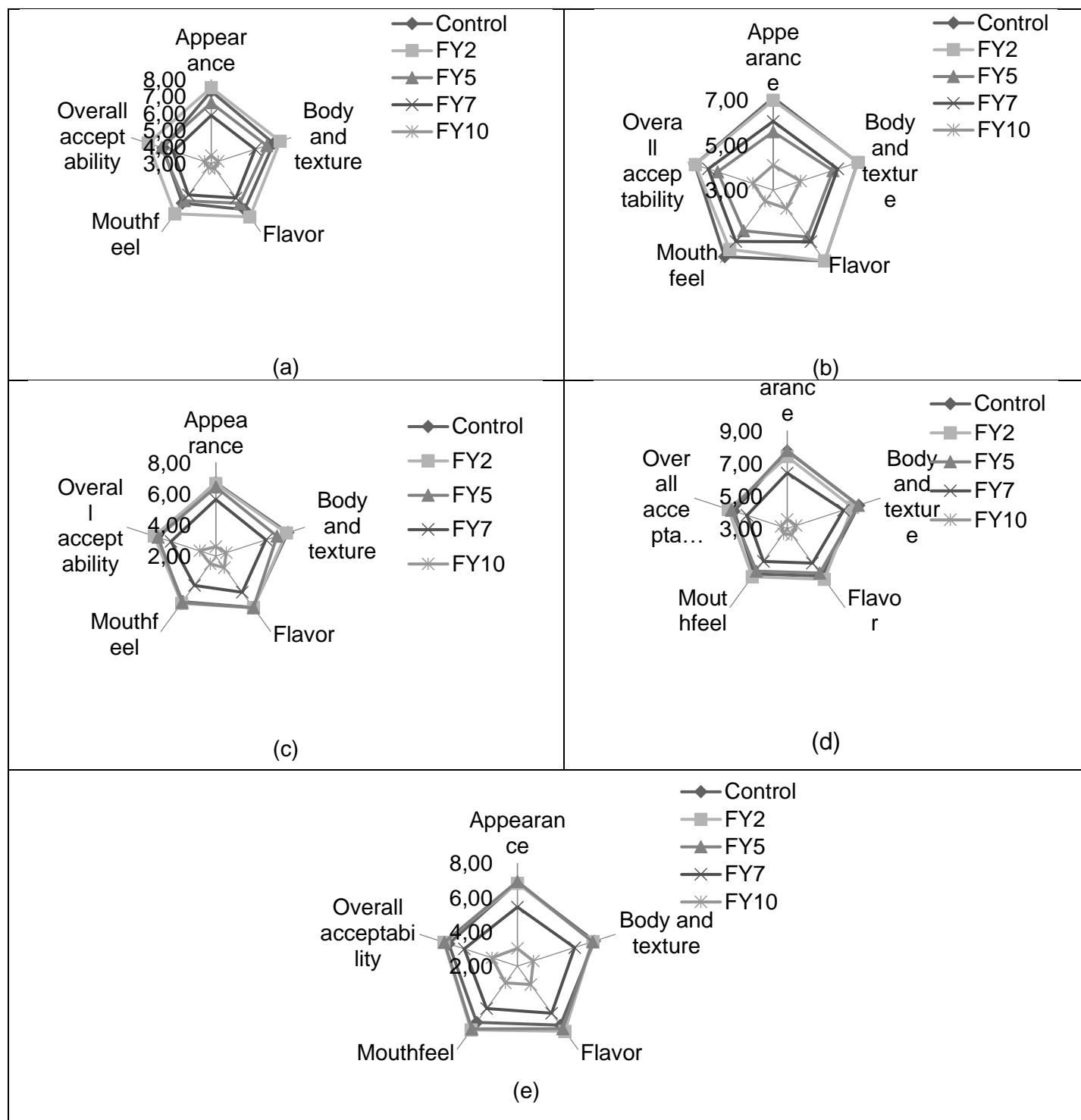


Figure 1. Sensory properties of yoghurt samples containing FSP on the 1st (a), 7th (b), 14th (c), 21st (d) and 28th (e) days of storage. (C: yoghurt without flaxseed powder, FY2: yoghurt containing 0.25% flaxseed powder, FY5: yoghurt containing 0.50% flaxseed powder, FY7: yoghurt containing 0.75% flaxseed powder, FY10: yoghurt containing 1% flaxseed powder.)

CONCLUSION

The addition of FSP affected significantly the acidity and pH values of yoghurt samples. The syneresis and WHC improved in yoghurt samples added FSP. L^* and WI values of samples decreased with increment of FSP concentration. In other colorimetric measurements, significant differences were determined with the addition of FSP. The viscosity (50 and 100 rpm) and K values indicated the increments depending FSP concentration. Flow behavior of all yoghurts was pseudoplastic. *L. bulgaricus* and *S. thermophilus* counts of samples were not affected by FSP. *L. bulgaricus*, *S. thermophilus*, molds-yeasts counts were in accordance with the Codex Alimentarius. The addition of FSP did not have any negative effect on yoghurt bacteria (*L. bulgaricus*, *S. thermophilus*). The sample containing 1% FSP (FY10) had the lowest syneresis, L^* , WI, viscosity (50 and 100 rpm), K values and sensory scores, and the highest WHC. Sensory scores by panelists to the samples C and FY2 were close. Opposition of obtained results in the previous studies using FSP in yoghurt should be less than 1%.

Funding: This research received no external funding.

Acknowledgments: This article is derived from the Master Thesis of Alperen Kalyas.

Conflicts of Interest: The authors declare no conflict of interest.

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