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INVESTIGATION OF USAGE OPPORTUNITIES OF DIFFERENT PROPORTIONS OF WHEY AND CHIA SEED FLOUR IN BUTTERMILK

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Abstract

Original scientific paper

In this study, it was aimed to obtain a functional beverage with high nutritional value by using whey and chia seed flour in the production of ayran. In addition to its high nutritional value, while preventing whey from causing environmental pollution as a waste material, it is aimed to obtain a new dietary product with chia seed flour containing 34-40g fiber per 100g, antioxidant, omega-3 fatty acid and rich protein content. Experimental ayran samples were prepared by adding whey and chia seed flour at different rates. K (Ayran), K_2 (Ayran + 1% Chia seed flour), K_3 (Ayran + 0,5% Chia seed flour , Group 1(Ayran (75%) + Whey (25%) + 1% Chia seed flour), Group 2 (Ayran (75%) + Whey (25%) + 0,5% Chia seed flour), Group 3 (Ayran (50%) + Whey (50%) + 1% Chia seed flour), Group 4 (Ayran (50%) + Whey (50%) + 0,5% Chia seed flour) were kept at +4 ℃ and their physical, chemical and sensory properties were examined on the 1st, 3rd, 5th, 7th and 14th days. In the experimental ayran samples we researched, the use of whey and chia seed flour was found to have a significant effect on psychrophilic bacteria count (log cfu/ml), coliform group bacteria (log cfu/ml), yeast-mold count (log cfu/ml), lactobacil spp. (log cfu/ml) and lactococ spp. (log cfu/ml), pH, acidity (lactic acid), water-soluble dry matter (%), serum separation (%), color values (L*, a*, b*) and sensory properties ($p<0,05$). In this experimental study, when microbiological, physical, chemical and sensory analyzes are taken into account, the groups that can be used in ayran production were determined as the K2 and K3 groups containing Chia seed flour and the 1st group containing 25% Whey + %1 Chia seed flour. The use of the 3rd and 4th experimental groups was not possible. It has been identified that these experimentally determined rates can provide a new product to the functional food industry by enabling the use of whey and chia seed flour.

Keywords: *Ayran, chia seed, physical, chemical and sensory analyses, whey.*

1 Introduction

In our society, which is becoming increasingly conscious about healthy nutrition, the tendency towards functional foods is increasing day by day in order to use natural resources more efficiently. The low consumption habits of milk and dairy products, which have a great importance in human nutrition and are included in the animal food class, and the short shelf life have made it necessary to consume most of the milk as fermented milk products. Yoghurt and ayran are the leading fermented milk products. Ayran contains all the nutritional properties of yoghurt at different rates depending on the amount of water added [1].

The majority of the residual material in the dairy industry occurs in cheese production. 70-90% of the milk used in cheese making is separated as whey. Whey can become an environmentally harmful product if it is disposed into the environment due to the proteins and other nutrients in its composition [1]. The chemicals formed as a result of fermentation of the organic substances contained in the whey discharged into the environment cause environmental pollution and pose a threat to living organisms [2]. In clinical studies, the use of whey in the treatment of cancer, AIDS, hepatitis B, cardiovascular diseases and osteoporosis has given successful results [3]. whey proteins are a superior source of essential amino acids than many proteins due to their biological properties and sulfur-containing amino acids such as cysteine and methionine. In the food industry, is added to many products such as confectionery, bakery products, meat products, soups, sauces, potato chips, snacks and various beverages [4,5].

Chia seed is a plant called '*Salvia hispanica* L.', which is used as food, medicine, cosmetics, perfumery and colouring matter [6]. Chia seed proteins have four different structures. These are globulin, albumin, gluten and prolamin. Globulin constitutes 52% of the total protein structure. The remaining part is shared equally by other proteins [7]. Approximately 1g of chia seed contains 0.75g of phenolic compounds such as chlorogenic acid, caffeic acid, quercetin and campferol in antioxidant structure. In addition, Chia seed, which is rich in calcium, phosphorus, potassium, magnesium, niacin, vitamin A and fibre content, is poor in sodium, iron, zinc and vitamin C [8]. Since 6% of Chia seed consists of soluble fibres,

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products with high dietary fibre content can be obtained [9]. Chia seed is used as a thickener in the food industry because it can absorb 27 times its weight in water and form mucilage [10]. It has been observed that it contributes to the nutritional value by being used in the production of many bakery products, especially bread, in many food products such as biscuits, ice cream and animal feeds.

The aim of this study was to investigate the effects of ayran production with different ratios of whey and Chia seed flour on microbiological, chemical and sensory properties of ayran. It was aimed to prevent economic losses, prevent environmental pollution and increase the nutritional value of ayran by using whey, which is a waste material in dairy industry. In addition, the product quality and shelf life of a new functional food production was investigated by taking advantage of the antioxidant, dietary pulp and appetite control properties of Chia seed containing 34-40g fibre per 100g.

2 Material Method

2.1 Material

The ayran and whey were obtained from Sivas Cumhuriyet University Food Studies Application and Research Centre, Chia seeds were obtained from a commercial company, chemicals were obtained from TEKKİM (Istanbul, Turkey) and media were obtained from BİOCAR (Istanbul, Turkey).

2.2 Method

The raw milk was wheyteurised and then produced with the addition of starter culture considering the ayran production process. Whey obtained from white cheese production was wheyteurised at 65℃ for 30 minutes and prepared for use. Chia seeds were ground into flour and then wheyteurised. The amount of chia seed flour and whey added was determined as a result of preliminary experiments and experimental samples were prepared. These are; K: Ayran (Control), K2: Ayran + 1% Chia seed flour, K3: Ayran + 0,5% Chia seed flour, 1: Ayran (75%) + whey (25%) + 1% Chia seed flour, 2: Ayran (75%) + whey (25%) + 0,5% Chia seed flour, 3: Ayran (50%) + whey (50%) + 1% Chia seed flour, 4: Ayran (50%) + whey (50%) + 0,5% Chia seed flour.

The prepared experimental samples were filled in the ayran filling machine, sealed and kept in cold storage at +4℃ on the 1st, 3rd, 5th, 7th and 14th days to be investigated in terms of physical, chemical, microbiological and sensory properties.

2.2.1 Microbiological analyses

Experimental ayran samples were inoculated on Violet Red Bile Agar medium for coliform group bacteria enumeration using the pouring plate method. It was left to incubation at 37℃ for 24 hours. For total psychrophilic bacteria group counting, Plate Count Agar medium was inoculated by pouring plate method. Colonies were counted by incubation at 6.5℃ for 10 days. In the enumeration of lactic acid bacteria in experimental ayran samples, appropriate distillations were sown on Man Rogosa Sharpe Agar medium for Lactobacillus species and M17 Agar medium for Lactococus species by pouring plate method. After sowing, incubation was performed at 37℃ for 48 hours. For total yeast and mould counts, Potato Dextrose Agar medium was sown by pouring plate method and colonies were counted by incubation at 25℃ for 4-5 days [11].

2.2.2 Physical and Chemical Analyses

The pH values of the experimental ayran samples and whey were measured with a digital pH meter (HANNA HI 98128) [12]. Titration acidity was determined by titrimetric method [13]. The water soluble dry matter content of experimental ayran samples and whey was measured by digital refractometer (MİLWAUKEE MA871) [14]. For the determination of serum separation; 25 ml of experimental samples (at 4±1℃) were added to the filter paper placed in a tared funnel and kept for 120 minutes. The amount of serum filtered into the tared flask was weighed and multiplied by 4 and expressed as % [15]. Colour analysis of the experimental ayran samples was carried out by calorimeter (MINOLTA CR-400) using CIELAB colour method. This technique gives numerical values of three colour scales (L*, a*, b*) and L* indicates whiteness and brightness, a* indicates red and green, and b* indicates yellow and blue [16].

2.2.3 Sensory Analyses

The experimental ayran samples were analysed by 8 panelists. Water and crackers were offered between different sample analyses to prevent the flavours from being affected by each other. Appearance and texture, odour and taste characteristics were taken into consideration and evaluated on a scale of 1-10 [17].

2.2.4 Statistical Analyses

IBM SPSS 23.0 package programme was used to evaluate the data obtained in this study. The differences between the experimental sample groups were determined by One-Way Anova test to evaluate all analysis data and statistically significant differences were determined by Tukey HSD (Honestly Significant Difference) test. The differences of the experimental ayran samples between days were determined by General Linear Model- Repeated Measures analysis, which is a repeated measures analysis, and the differences obtained were determined by Bonferroni test [18].

3 Results and Discussion

3.1 Microbiological Analysis Results of Experimental Samples

According to the Turkish Food Codex Communiqué on Fermented Milk Products (Communiqué no: 2009/25), ayran should contain at least 10^6 total specific microorganisms (cfu/ml) [19]. Except for yeast, which is used probiotic in ayran, it should be between 10^2-10^3 in 2 out of 5 samples and less than 10^2 in 3 samples. Mould

should be between 10^2-10^3 in 2 out of 5 samples and less than 10^2 in 3 samples, and E. coli and other pathogenic microorganisms should not be present [20]. Microbiological analysis findings of the experimental samples are given in Table 1.

The number of lactic acid bacteria in whey used in the experimental samples was determined as 1.01 log cfu/ml. Coliform bacteria and yeast-mould counts were found below the detectable value (< 10 cfu/ml).

In the experimental ayran samples, the number of coliform bacteria was found below the detectable level during storage. Bekiş (2019), found the number of coliform bacteria below the detectable level in his thesis study investigating the possibilities of using whey in kefir [17]. Ünalan (2022), stated that the number of coliform bacteria was below the detectable level in his thesis study investigating the possibilities of using different ratios of whey and mint oil in ayran [21]. Tamuçay-Özünlü and Koçak (2010) determined the number of coliform bacteria below the detectable level in their study in which they investigated the ayran quality of different heat treatment applications to milk [22].

Total psychrophilic bacteria counts of the experimental ayran samples were found in the range of 3.12-5.38 log cfu/ml. When the results of intra-group inter-day analyses of the experimental ayran samples during the storage period were examined, the difference between the K and K2 experimental ayran samples on the 1st and 3rd days and on the 5th, 7th and 14th days was found to be statistically significant ($p<0.05$).When the experimental ayran samples were evaluated between groups during the storage period, the difference between the K3 and 1st groups on the 1st and 3rd days and between the K, K2, 2nd, 3rd and 4th groups was not found to be statistically significant ($p>0.05$). When the results of the analysis were evaluated, the presence of whey and Chia seed flour did not affect the total number of psychrophilic bacteria due to the preparation of experimental samples by wheyteurising whey, ayran and Chia seed flour. It is thought that the existing differences are due to the packaging during the shelf life of ayran. Doğan (2022), in his study investigating the production of probiotic beverage by adding black cumin seed oil to whey and pomegranate juice mixture, stated that the total number of psychrophilic bacteria was in the range of 2.08-6.57 log cfu/ml [23]. Akarca and Tomar (2019), in their study on the chemical and microbiological properties of strained (pouch) yoghurt sold in the district markets of Afyonkarahisar province, stated that the total number of psychrophilic bacteria was found in the range of 2.50-4.39 log cfu/g [24]. Ünalan (2022) reported that the total number of psychrophilic bacteria was found in the range of 3.23-5.32 log cfu/ml in his thesis study in which he investigated the possibilities of using different ratios of whey and peppermint oil in ayran [21]. According to our research findings, the total number of psychrophile organisms was found close to these studies.

The number of lactobacil spp. was found to be in the range of 2.53-3.23 log cfu/ml during the storage period of the experimental ayran samples. The difference between the experimental ayran samples was found to be insignificant ($p>0.05$). It is thought that the presence of chia seed flour and whey did not affect the number of lactobacil spp.

Tulukoğlu (2019) found the number of Lactobacil spp. in the range of 8.70-13.11 log cfu/ml in a study investigating the use of whey in the production of Izmir tulum cheese [25]. Ünalan (2022) found the number of Lactobacil spp. in the range of 2.34-3.40 log cfu/ml in his thesis study investigating the possibilities of using different ratios of whey and mint oil in ayran [21]. Our research findings were close to the findings of Ünalan (2022) and lower than the findings of Tulukoğlu (2019) [21, 25]. The number of lactococ spp. in the experimental samples was found to be in the range of 2.70-3.53 log cfu/ml. The difference between the days within the group during the storage period was found to be statistically insignificant ($p>0.05$). The difference between K, K2 and K3 groups was not statistically significant ($p>0.05$). The difference between groups K2, K3 containing chia seed flour and groups 1, 2, 3, and 4 containing whey was statistically significant ($p<0.05$). It is thought that the presence of whey decreased the number of lactococ spp. Tulukoğlu (2019), in a study investigating the use of whey culture in the production of Izmir tulum cheese, found the number of Lactococ spp. in the range of 6.77-10.46 log cfu/ml [25]. Dinç and Kahyaoğlu (2021), in their study on the examination of microbiological and physicochemical properties of commercial ayran, found that the number of Lactococ spp. number in the range of 2.12-3.66 log cfu/ml, Ünalan (2022) found the number of Lactococ spp. in the range of 2.59-4.15 log cfu/ml in his thesis study investigating the possibilities of using different ratios of whey and mint oil in separation [21, 26]. While our research findings were lower than the findings of Tulukoğlu (2019), they were close to the findings of Ünalan (2022) and Dinç and Kahyaoğlu (2021) [21, 25, 26].

In this study, the differences between the groups in yeast-mould count were not statistically significant ($p>0.05$). The difference between days 1, 3 and 5, 7, 14 was found to be statistically significant ($p<0.05$).

The results on days 5, 7 and 14 were found to be appropriate according to the Turkish Food Codex Communiqué on Fermented Milk Products (Communiqué No: 2009/25) [19]. Due to the wheyteurisation of the experimental ayran samples, it is thought that the yeast-mould count was below the detectable level on days 1 and 3. It is thought that whey and Chia seed flour do not affect the product quality negatively and the increase in yeast-mould count is related to shelf life.

Hayatoğlu (2021) determined the yeast-mould count in the range of 2.10-2.94 log cfu/ml in a study investigating the physical, chemical and microbiological properties of ayran produced with the addition of probiotic bacteria [27]. Akçay (2016) determined the yeast-mould count in the range of 2.36-4.71 log cfu/ml in a study on the production of spicy ayran and investigation of some of its properties [28]. Tamuçay-Özünlü and Koçak (2010), in their study in which they examined the effect of different heat treatment applications on ayran quality, stated that they found the yeast-mould count below the detectable level [22]. Çelik et al. (2016), in their study investigating the physicochemical and microbiological quality of yoghurt and ayran produced in Kırklareli, found the yeast-

mould count in the range of $2.3x10^1$-$1.9x10^5$ log cfu/ml and stated that it was above the limit value [29]. It was determined that our research findings were close to the findings of Akçay (2016) and different from the findings of other studies.

3.2 Chemical Analysis Results of Experimental Samples

Chemical analyses of the experimental samples are given in Table 2. Whey pH value was 6.31, % dry matter was 5.75 and titration acidity was 0.11 in terms of lactic acid.

L* value indicates whiteness and brightness. The results of within-group, between-day and between-group analyses of the experimental samples were found to be statistically significant ($p<0.05$). When the results between the groups were analysed, it was seen that the L* values of the 1st, 2nd, 3rd and 4th groups containing whey decreased compared to the K2, K3 groups containing K and Chia seed flour. In our study, it is thought that whey-containing samples reduce the brightness. Temen (2018), in a study investigating the production and some properties of quinoa flour-added ayran, L* value was reported in the range of 84.43-85.96 [29]. Ürkek et al. (2021), in a study investigating the effect of chia addition on the physicochemical and sensory properties of ice cream, reported that they found the L* value in the range of 58.98-80.30 [30]. While our research findings were close to the findings of Temen (2018), they were higher than the findings of Ürkek et al. (2021) [29, 30]. Because, our study was used whey.

When the results of the analysis between the groups were evaluated, it was determined that the a* value of the 1st, 2nd, 3rd and 4th samples containing whey was lower than the K, K2 and K3 groups. Since whey is a yellowish-green coloured organic liquid, it is seen that the greenness value increases as the whey ratio increases. It is thought that chia seed flour adversely affects the homogeneous distribution due to its particulate structure and makes a difference in the findings during storage. Temen (2018) determined the a* value in the range of -2.92 to -2.67 in a study investigating the production and some properties of ayran with quinoa flour addition [29]. Ürkek et al. (2021) reported the a* value in the range of -2.86 to 1.54 in a study investigating the effect of chia addition on the physicochemical and sensory properties of ice cream [30]. It was determined that our research findings were not close to these studies.

When the b* value is evaluated between the groups, the b* value increases in groups 1, 2, 3 and 4 containing whey compared to K2 and K3 groups containing K and Chia seed flour ($p<0.05$). It is thought that the whey ratio increases the yellowness. Temen (2018), in a study investigating the production and some properties of quinoa flour-added ayran, determined the b* value in the range of 7.49 to 7.90 [29]. Ürkek et al. (2021), in a study investigating the effect of chia addition on the physicochemical and sensory properties of ice cream, determined the b* value in the range of 0.31-6.81. It was determined that our research findings were not close to these studies [30].

When the experimental samples were evaluated in terms of pH and titration acidity, the statistical difference was found to be insignificant ($p>0.05$, table 2). pH value was determined between 4.05 and 4.78. Titration acidity was determined insignificant when statistical analyzis (Table 2).

When the water soluble dry matter values of the experimental ayran samples were analysed during the storage period, it was found that the water soluble dry matter value increased as the % whey ratio increased and this is thought to be due to the high dry matter content of whey.

When the serum separation values of the samples during the preservation period were compared with the 1st, 2nd, 3rd and 4th groups and K, K2 and K3 groups, the difference was found to be statistically significant ($p<0.05$). It was determined that the presence of whey increased the serum separation value.

3.3 Sensory Analysis Results of Experimental Samples

The sensory analysis findings of the experimental samples are given in Table 3. When the samples were analysed in terms of appearance, the highest score was obtained by group K3 and the lowest score was obtained by group 3 containing 50% whey. It was observed that the homogenous appearance decreased as the storage time and whey amount increased. K2 and K3 groups received the highest score in the pleasant and distinctive odour parameter during the storage period. 3rd and 4th groups containing 50% whey received the lowest score. It is thought that there is a decrease in pleasant odour due to the increase in whey ratio. During the storage period of the experimental ayran samples, the highest score for the pleasant and distinctive flavour parameter was obtained by the K2 group and the lowest score was obtained by the 4th group. It is thought that the taste of the panellists decreased with the increase in sour taste as the storage time and whey amount increased. In terms of sensory analyses, it was determined that K2 and K3 groups received the highest score by the panelists during the storage period. It is thought that chia seed flour provided sufficient flavour and consistency among the reasons why K3 group was liked. Among the experimental ayran groups prepared with whey, it was determined that the 3rd and 4th groups received the lowest scores in all parameters. The increase in acidity and serum separation during storage shortens the shelf life of ayran. The reason for the 3rd and 4th experimental groups to get the lowest score is thought to be that whey accelerates acid formation. Bekiş (2019), in his study investigating the possibilities of using whey in kefir, stated that the homogeneous and foamy structure was lost in the last days of storage, the viscosity decreased and the appearance characteristics were less liked by the panellists due to reasons such as serum separation [17].

In this study we conducted, microbiological, physical, chemical and considering the sensory analyses, it may be possible to use in milk production. Groups K2 and K3 containing Chia seed flour and 25% Whey + 10 g Chia seeds. It was determined as the 1st group containing flour. These experimentally determined rates functional food by allowing the use of whey and chia seed flour. It has been determined that it can bring a new product to the industry.

Table 1. Microbiological analysis results of experimental ayran samples.

Analysis	Groups	Storage time (days)				
		1	3	5	7	14
TPAB	K	4,52±0,01^{bA}	4,68±0,02^{bA}	5,31±0,02^{cB}	5,34±0,03^{bB}	5,38±0,10^{aB}
	K2	4,28±0,02^{bA}	4,32±0,01^{bA}	4,93±0,02^{bB}	5,12±0,08^{bB}	5,13±0,05^{aB}
	K3	3,12±0,02^{aA}	3,47±0,03^{aA}	3,77±0,14^{aA}	5,04±0,07^{bB}	5,21±0,18^{aB}
	1	3,56±0,02^{aA}	3,83±0,02^{aA}	4,45±0,07^{bB}	5,04±0,08^{bC}	5,07±0,05^{aC}
	2	4,42±0,02^{bA}	4,73±0,03^{bA}	5,24±0,02^{cB}	4,88±0,01^{aA}	5,15±0,30^{aB}
	3	4,08±0,01^{bA}	4,66±0,02^{bB}	5,17±0,02^{cC}	5,03±0,03^{bC}	5,09±0,08^{aC}
	4	4,35±0,02^{bA}	4,92±0,02^{bB}	5,00±0,08^{cB}	5,03±0,04^{bB}	5,07±0,03^{aB}
Lactobacil	K	3,23±0,01	3,15±0,01	2,92±0,01	3,06±0,01	2,89±0,02
	K2	2,85±0,02	2,76±0,05	2,69±0,01	2,68±0,02	2,79±0,01
	K3	2,87±0,03	2,78±0,02	2,68±0,01	2,71±0,02	2,81±0,01
	1	2,83±0,01	2,77±0,01	2,69±0,02	2,65±0,02	2,90±0,01
	2	2,93±0,01	2,83±0,01	2,79±0,01	2,67±0,02	2,90±0,01
	3	2,72±0,01	2,77±0,06	2,84±0,01	2,53±0,01	2,88±0,01
	4	2,73±0,02	2,92±0,01	2,67±0,01	2,65±0,02	2,90±0,01
Lactococ	K	3,53±0,01^{b}	3,51±0,01^{b}	3,19±0,01^{b}	3,25±0,01^{b}	3,11±0,01^{b}
	K2	3,08±0,02^{b}	2,83±0,01^{a}	2,79±0,02^{a}	2,86±0,01^{a}	3,23±0,01^{b}
	K3	3,06±0,01^{b}	2,82±0,01^{a}	2,78±0,02^{a}	2,90±0,01^{a}	3,21±0,01^{b}
	1	2,77±0,13^{a}	2,78±0,02^{a}	2,80±0,04^{a}	2,82±0,02^{a}	2,90±0,02^{a}
	2	2,77±0,10^{a}	2,78±0,27^{a}	2,70±0,12^{a}	2,71±0,12^{a}	2,85±0,03^{a}
	3	2,81±0,02^{a}	2,79±0,01^{a}	2,79±0,01^{a}	2,78±0,01^{a}	2,91±0,01^{a}
	4	2,92±0,01^{a}	2,90±0,02^{a}	2,85±0,01^{a}	2,79±0,01^{a}	2,91±0,01^{a}
Mould-yeast	K	<1	<1	4,30±0,01	4,40±0,01	4,65±0,02
	K2	<1	<1	4,33±0,03	4,75±0,02	4,96±0,02
	K3	<1	<1	4,18±0,02	4,54±0,03	4,63±0,28
	1	<1	<1	4,49±0,02	4,73±0,06	4,86±0,04
	2	<1	<1	4,43±0,02	4,52±0,02	4,83±0,01
	3	<1	<1	4,32±0,01	4,61±0,02	4,74±0,01
	4	<1	<1	4,26±0,02	4,48±0,01	4,62±0,02

**K:Control, K2: Ayran + 1% Chia seed flour, K3: Ayran + 0,5% Chia seed flour, 1: Ayran (75%) + WHEY (25%) + 1% Chia seed flour, 2: Ayran (75%) + WHEY (25%) + 0,5% Chia seed flour, 3: Ayran (50%) + WHEY (50%) + 1% Chia seed flour, 4: Ayran (50%) + WHEY (50%) + 0,5% Chia seed flour. a-g: The difference between samples with different letters in the same column is significant (p<0,05). A-C: The difference between samples with different letters in the same line is significant (p<0,05). The not difference between samples with different letters in the same line is insignificant and not determinad (p>0,05)

Table 2. Chemical analysis results of experimental ayran samples.

Analysis	Groups	Storage time (days)				
		1	3	5	7	14
*L** value	K	91,26±0,68^{eA}	92,76±0,34^{cB}	94,75±0,58^{cC}	97,16±0,19^{dD}	98,95±1,20^{cE}
	K2	90,86±1,46^{dA}	93,35±0,11^{dB}	94,98±0,54^{cC}	96,03±0,25^{cD}	98,31±0,02^{cE}
	K3	91,79±0,05^{eA}	93,49±0,54^{eB}	96,82±0,49^{dC}	98,39±0,02^{eD}	99,57±0,12^{dE}
	1	87,95±0,06^{bA}	90,86±0,55^{bB}	92,58±0,72^{bC}	94,63±0,12^{bD}	96,12±0,22^{bE}
	2	88,71±0,02^{cA}	90,56±0,80^{bB}	92,68±0,93^{bC}	94,35±1,13^{bD}	96,60±0,09^{bE}
	3	86,22±0,10^{aA}	88,98±0,54^{aB}	90,87±0,75^{aC}	93,73±0,11^{aD}	96,58±0,30^{bE}
	4	86,35±0,09^{aA}	88,69±1,41^{aB}	90,31±1,22^{aC}	93,55±1,45^{aD}	95,08±0,45^{aE}
*a** value	K	-0,60±0,01^{bA}	-0,76±0,01^{bA}	-0,82±0,09^{bA}	-0,96±0,03^{bA}	-2,87±0,09^{aB}
	K1	-0,48±0,03^{bA}	-1,80±0,02^{cB}	-1,86±0,10^{cB}	-1,92±0,03^{cB}	-3,51±0,10^{aC}
	K2	-0,40±0,03^{bA}	-1,16±0,02^{cB}	-1,29±0,12^{cB}	-1,51±0,01^{cB}	-3,11±0,12^{aC}
	1	-1,10±0,02^{aA}	-2,10±0,01^{aB}	-2,63±0,14^{aB}	-2,69±0,03^{aB}	-3,22±0,26^{aC}
	2	-1,18±0,02^{aA}	-2,30±0,01^{aB}	-2,42±0,05^{aB}	-2,65±1,19^{aB}	-3,34±0,15^{aC}
	3	-1,77±0,06^{aA}	-2,58±0,05^{aB}	-2,76±0,09^{aB}	-2,82±0,04^{aB}	-3,80±0,46^{aC}
	4	-1,98±0,05^{aA}	-2,78±0,02^{aB}	-2,92±0,09^{aB}	-2,95±0,16^{aB}	-3,95±0,75^{aC}
*b** value	K	3,89±0,09^{aE}	2,75±0,06^{aD}	1,61±0,08^{aC}	0,26±0,11^{aB}	-2,31±0,05^{bA}
	K1	4,94±0,04^{bE}	3,82±0,07^{bD}	2,06±0,32^{bC}	0,68±0,01^{aB}	-2,95±0,09^{bA}
	K2	4,86±0,02^{bE}	3,77±0,14^{bD}	2,59±0,07^{bC}	1,09±0,01^{bB}	-2,81±0,19^{bA}
	1	5,03±0,03^{cE}	3,96±0,32^{bD}	2,85±0,08^{bC}	1,20±0,08^{bB}	-3,44±0,38^{sA}
	2	5,01±0,05^{cD}	3,61±0,69^{bC}	2,99±0,10^{bB}	2,27±0,59^{cB}	-3,91±0,35^{sA}
	3	5,13±0,04^{cE}	4,88±0,14^{cD}	3,17±0,59^{cC}	1,80±0,08^{cB}	-3,40±0,14^{sA}
	4	5,25±0,02^{cE}	4,58±0,04^{cD}	3,79±0,17^{cC}	1,85±0,59^{cB}	-3,63±0,09^{aA}
pH	K	4,18±0,00	4,09±0,00	4,11±0,00	4,10±0,00	4,05±0,01
	K1	4,17±0,00	4,13±0,00	4,11±0,01	4,09±0,00	4,05±0,00
	K2	4,16±0,01	4,12±0,00	4,07±0,00	4,07±0,00	4,05±0,00
	1	4,40±0,00	4,31±0,00	4,26±0,00	4,33±0,01	4,45±0,00
	2	4,41±0,01	4,28±0,01	4,27±0,01	4,21±0,01	4,23±0,01
	3	4,78±0,00	4,59±0,00	4,46±0,00	4,58±0,00	4,55±0,00
	4	4,76±0,00	4,58±0,01	4,48±0,00	4,52±0,01	4,54±0,01

Table 2 (Continued). Chemical analysis results of experimental ayran samples.

Acidity	K	0,49±0,001	0,54±0,003	0,46±0,001	0,51±0,001	0,45±0,080
	K1	0,47±0,002	0,46±0,001	0,43±0,001	0,47±0,002	0,47±0,010
	K2	0,46±0,001	0,45±0,011	0,41±0,002	0,48±0,000	0,41±0,030
	1	0,57±0,001	0,57±0,001	0,50±0,000	0,55±0,001	0,59±0,120
	2	0,53±0,001	0,55±0,004	0,51±0,010	0,59±0,002	0,56±0,080
	3	0,62±0,001	0,63±0,001	0,64±0,001	0,66±0,000	0,65±0,130
	4	0,61±0,001	0,64±0,000	0,60±0,001	0,62±0,000	0,64±0,050
Dry matter	K	$2,25±0,00^{aB}$	$2,63±0,04^{aB}$	$2,20±0,00^{aB}$	$1,07±0,02^{aA}$	$0,96±0,01^{aA}$
	K1	$3,93±1,03^{bC}$	$3,82±0,00^{bC}$	$2,76±0,04^{aB}$	$2,23±0,01^{bB}$	$0,88±0,11^{aA}$
	K2	$3,88±0,04^{bB}$	$3,20±0,10^{bB}$	$3,18±0,04^{bB}$	$2,53±0,11^{bA}$	$2,53±0,04^{bA}$
	1	$3,80±0,00^{bB}$	$3,88±0,74^{bB}$	$3,60±0,10^{bB}$	$3,08±0,04^{cB}$	$2,60±0,00^{bA}$
	2	$3,88±0,01^{bB}$	$3,86±0,04^{bB}$	$3,85±0,04^{bB}$	$3,50±0,64^{cB}$	$2,65±0,00^{bA}$
	3	$3,96±0,01^{bA}$	$3,93±0,04^{bA}$	$3,80±0,00^{bA}$	$3,76±0,01^{cA}$	$3,70±0,07^{cA}$
	4	$4,80±0,01^{cB}$	$4,46±0,10^{cB}$	$3,73±0,05^{bA}$	$3,38±0,74^{cA}$	$3,34±0,04^{cA}$
Serum seperation	K	$69,63±2,02^{bA}$	$70,65±0,10^{bA}$	$70,15±0,10^{bA}$	$71,40±0,00^{cA}$	$72,83±0,00^{dA}$
	K1	$66,22±0,00^{aB}$	$66,18±0,01^{aB}$	$66,03±0,04^{aB}$	$66,45±0,10^{aB}$	$64,25±0,10^{aA}$
	K2	$67,41±0,11^{aB}$	$66,89±0,04^{aB}$	$66,78±0,04^{aB}$	$66,63±0,04^{aB}$	$65,40±0,00a^{A}$
	1	$68,21±0,04^{aC}$	$67,66±0,02^{aC}$	$66,23±0,04^{aB}$	$66,81±0,01^{aB}$	$64,70±0,42^{aA}$
	2	$73,13±0,11^{dC}$	$72,45±0,35^{cC}$	$71,98±0,04^{cB}$	$71,86±0,10^{cB}$	$70,68±0,11^{cA}$
	3	$71,00±0,00^{cC}$	$70,58±0,11^{bC}$	$69,45±0,10^{bB}$	$69,41±0,01^{bB}$	$68,00±0,00^{bA}$
	4	$75,20±0,00^{eC}$	$74,03±0,04^{dC}$	$73,80±0,00^{dB}$	$73,52±0,04^{dB}$	$72,48±0,95^{dA}$

**K:Control, K2: Ayran + 1% Chia seed flour, K3: Ayran + 0,5% Chia seed flour, 1: Ayran (75%) + WHEY (25%) + 1% Chia seed flour, 2: Ayran (75%) + WHEY (25%) + 0,5% Chia seed flour, 3: Ayran (50%) + WHEY (50%) + 1% Chia seed flour, 4: Ayran (50%) + WHEY (50%) + 0,5% Chia seed flour. a-g: The difference between samples with different letters in the same column is significant ($p<0,05$). A-C: The difference between samples with different letters in the same line is significant ($p<0,05$). The not difference between samples with different letters in the same line is insignificant and not determinad ($p>0,05$)

Table 3. Sensory analysis total score results of experimental ayran samples.

Analysis	Groups						
	K	K2	K3	1	2	3	4
Homogeneity	45,94	37,18	37,18	37,18	37,18	37,18	37,18
Colour	45,56	45,56	45,56	45,56	45,56	45,56	45,56
Saturation	38,94	38,94	38,94	38,94	38,94	38,94	38,94
Vizkozity	37,82	37,82	37,82	37,82	37,82	37,82	37,82
Serum Seperation	17,19	17,19	17,19	17,19	17,19	17,19	17,19
Odour	34,44	34,44	34,44	34,44	34,44	34,44	34,44

**K:Control, K2: Ayran + 1% Chia seed flour, K3: Ayran + 0,5% Chia seed flour, 1: Ayran (75%) + WHEY (25%) + 1% Chia seed flour, 2: Ayran (75%) + WHEY (25%) + 0,5% Chia seed flour, 3: Ayran (50%) + WHEY (50%) + 1% Chia seed flour, 4: Ayran (50%) + WHEY (50%) + 0,5% Chia seed flour. a-g: The difference between samples with different letters in the same column is significant ($p<0,05$). A-C: The difference between samples with different letters in the same line is significant ($p<0,05$).

Declaration

Ethics committee approval is not required.

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