# The effect of plant extracts on antioxidant potential, microbial and sensory attributes of stirred yoghurt

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## Abstract

The aim of this study was to use various plants to increase the functional properties of yoghurt. The ethanol extracts of three different plant (Mentha piperita L., Ocimum basilicum L., and Hibiscus sabdariffa L.) extracts were added to the stirred type yoghurts at different ratios (0.1 %, 0.3 % and 0.5 %). The pH values, colour values, antioxidant activity, microbial and sensory attributes of yoghurt were evaluated on the storage days of 0, 7, 14, 21, and 28. The lowest pH values were found at the beginning (4.29) and at the end (3.95) of the storage period in samples containing 0.5 % hibiscus extract. The addition of plant extracts in amounts of 0.3 % and higher decreased the lightness ( $L^*$ ) value of yoghurt (p<0.05), and the maximum decrease was found in yoghurt samples containing 0.5% hibiscus ethanol extract. The concentrations of added mint and basil extracts increased the yellowness ( $b^*$ ) value (p<0.05), while the addition of hibiscus extract significantly increased the redness value (p<0.05). Hibiscus ethanol extract significantly reduced the growth of total aerobic mesophilic bacteria (TAMB), lactic acid bacteria (LAB) and Lactococcus/Streptococcus bacteria (p<0.05). The antioxidant activity of all samples increased with adding plant extracts. According to the results of sensory analysis, the addition of 0.1 % plant extracts to yoghurts rated higher scores than that of the control sample, but the addition of 0.3 % and 0.5 %ethanol extracts negatively affected the sensory properties. It can be concluded that adding plant extracts had a positive effect on the sensory and functional properties of yoghurt.

#### Key words: yoghurt; plant extract; antioxidant potential; colour

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### Introduction

Yoghurt is the most widely consumed fermented dairy product in the world today. It has a rich content of protein, omega 3 fatty acids, vitamins and minerals (Gao et al., 2018; Paszczyk et al., 2020). Although yoghurt is usually produced by fermentation with *Streptococcus thermophilus* and *Lactobacillus delbrueckii* spp. *bulgaricus* species bacteria, various starter cultures can be used in yoghurt the production (Freitas, 2017; Yao et al., 2017; Senadeera et al., 2018; Vieira et al., 2019).

Many different types of yoghurt are produced industrially. The most well-known of these types are stirred, set-style and liquid yoghurt. Also, each type is divided into full-fat, half-fat and non-fat classes according to the amount of milk fat it contains (Chandan and O'Rell, 2013).

Plants are known to have significant effects on consumer health, including antioxidants, antimicrobial, anti-inflammatory and anti-carcinogenic (Tapsell et al., 2006; Akarca et al., 2019). Mint (Mentha spp.) is an aromatic plant that grows widely in the temperate regions of the northern hemisphere (Soilhi et al., 2019). Mint is an important medicinal and aromatic plant and the genus contains between 25 and 30 species (Gupta et al., 2017). Basil (Ocimum basilicum L.) is a member of the Lamiaceae family and is a one-year herbaceous plant originating in Southeast Asia. The economic value of the plant, which can grow indigenously as well as produced by cultivation, is quite high (Varga et al., 2017). Hibiscus (Hibiscus sabdariffa L.) is a plant belonging to the Malvaceae family that grows in tropical and sub-tropical regions and there are about 300 species in the genus (Wang et al., 2012; Akarca 2019). The plant, which has an extremely high economic value, is used for many different purposes (carbonated / non-carbonated beverage, food colouring, food additives, etc.) (Hervert-Hernández and Goni, 2012; Sindi et al., 2014).

As consumers health concerns have increased in recent years, food producers' tendency to produce functional food products has increased as well (Illupapalayam et al., 2014). In recent years, interest in yoghurt as a functional food has increased greatly. Yoghurt has been added to various ingredients in order to enhance therapeutic, nutritional and sensory properties (Singh et al., 2011; Adegoke et al., 2013; Illupapalayam et al., 2014). For purpose of increasing the sensory and functional properties of yoghurt, various plants have been added (Singh et al., 2011; Adegoke et al., 2013; Illupapalayam et al., 2014)., Kumar et al. (2013) added peppermint leaves to yoghurts at various rates (2 %, 4 %, and 6 %) and reported that 2 % participation positively affected sensory values of the samples and extended the shelf life of the samples by 10 days. In another study, Halal and Tagliazucchi (2018) reported that the addition of cinnamon powder to yoghurts increased the total phenolic content and radical scavenging activity. Amirdivani and Baba (2011) stated that the participation of the addition of herbal extracts to yoghurts increases the acidity of yoghurts by improving the activity of yoghurt fermentation bacteria. The researchers reported that yoghurt bacteria had higher proteolytic activity in yoghurts with peppermint, dill and basil added. Gurkan and Hayaloglu (2017), on the other hand, found that the added basil powder and basil water extract in yoghurts improved sensory properties of the samples and increased its' volatile compounds content. Kim et al. (2019) investigated the sensory properties of hibiscus added to milk, kefir and yoghurts. They reported that the highest sensory scores were obtained for 1 % hibiscus added milk samples aa well as for 2 % and 3 % Hibiscus added yoghurt samples.

In some previous studies, mint, basil and hibiscus extracts were added to various dairy products alone. There is no study yet focused on comparing the effects of these three plant extracts to yoghurts and comparing their effects on various properties of yoghurts. Therefore, the main aim of the present study was to produce yoghurt containing ethanol extracts of three different plants (mint, basil and hibiscus) and investigate the effect of these extracts on the acidity, colour values, microbial stability, sensory properties and potential health effect by testing antioxidant activity.

## Materials and method

#### Material

Cow milk used in the production of yoghurt is provided from one of the dairy producers in Afyonkarahisar while the plants were provided from the Cakıroglu Spice Inc. (Afyonkarahisar, Turkey) company.

#### Production of yoghurts

Production of yoghurts was carried out according to Senel et al. (2006), with some modifications. Cow milk (°SH 7.1, pH 6.4, fat 3.2 5% protein 3.08 %, dry matter 9.75 % and density 1.028 g/mL was pasteurized at 85 °C for 20 min.) and cooled down to the incubation temperature (47 °C). Then, Streptococcus thermophilus (Cryofast ST051, Italy) and Lactobacillus delbrueckii spp. bulgaricus (Lyofast SP5, Italy) cultures were inoculated and mixed for 5 min. Inoculated samples were poured into 200 g packages and then left to fermentation at 47 °C. Subsequently plant ethanol extracts of Mentha piperita L. (Mint), Ocimum basilicum L. (Basil) and Hibiscus sabdariffa L. (Hibiscus)) were added separately (0.1 %, 0.3 %, and 0.5 %) and the containers were sealed and stored at +4 °C for 28 days.

#### Preparation of plant extracts

Extraction of used plants in the study was carried out according to Wei et al. (2011), with some modifications. The plants were first ground in a laboratory-type mill. Then, 100 g of powdered plants were weighed, 400 mL of 80 % ethyl alcohol (1:3 w/v) was added and mixtures were mixed in the shaker (Wiseshake SHO2D, Witeg, Germany) for 24 hours at 120 rpm with the mixture. At the end of the mixing process, the mixture was filtered through a sterilized 22-mm-pore filter paper (Whatman No. 22). The filtrate was then taken to a rotary evaporator (Heidolph Hei-VAP value, Germany) and the alcohol and extract were separated from each other at 100 rpm at 60 °C.

#### Physical analyses

The pH values of yoghurt samples were carried out using Ohaus (ST 5000, USA) while colour analyses (L\*, a\* and b\*) were carried out using a colorimeter (Minolta Chroma Meter CR-400, Osaka) according to Cappato et al (2018).

#### Determination of antioxidant activity

Antioxidant activity of samples were determined by the DPPH (1,1-diphenyl-2-picrylhydrazyl radical) method. The yoghurt samples were then left at 45  $^{\circ}$ C

for 10 minutes and then centrifuged (10 000 rpm, 10 min, at 4 °C). Supernatant was taken and the pH was adjusted to 7.0 using 0.1 M NaOH. The neutralized supernatant was centrifuged again (10 000 rpm, 10 min, at 4 °C). The supernatant obtained was then used for analysis. 10 g of yoghurt sample was mixed with 2.5 ml of distilled water and 1 mL of 0.1 M HCL was added to the mixture and the pH was adjusted to 4.0. For the determination of antioxidant activity, 250 µL of yoghurt extract was added into 3 mL of 60 µL in ethanol DPPH (0.0023 g DPPH in 100 mL of 96 % ethanol). The mixture was shacked and incubated at room temperature in a dark environment for about 30 seconds. Then the absorbance was read at 517 nm. The inhibition percent was calculated as follows (Apostolidis et al., 2007; Jung et al, 2016).

DPPH scavenging activity (%) =  $1 - (A_{sample} / A_{control}) \cdot 100$ 

#### Microbiological analysis

The total aerobic mesophilic bacteria (TAMB), lactic acid bacteria (LAB), *Lactococcus/Streptococcus* bacteria, lipolytic and proteolytic bacteria counts in yoghurt samples were determined by the spread plate method (ISO, 1999).

TAMB counts analysis was performed using the Plate Count Agar (Merck, Germany, 1.05463) (PCA) media. Cultivated Petri dishes were allowed to incubate at 30 °C for 48-72 h under aerobic conditions (ISO, 2013a; ISO, 2013b). For the enumeration of lactic acid bacteria, Man Rogasa and Sharpe Agar (MRS) (Merck, Germany, 110660) media were used. Accordingly, Petri dishes were incubated in a jar (Merck 1.16387) under anaerobic conditions at 30 °C for 24-48 hours (Kneifel and Berger, 1994). Lactococcus/Streptococcus spp. counts were determined using M-17 Agar (Merck, Germany 1.15108) medium. The Petri dishes were incubated at 30 °C for 24-48 hours under aerobic conditions (Corroler et al., 1998). Tributyrin Agar (Sigma-Aldrich, USA, 91015) and Plate Count Skimmed Milk Agar (Merck, Germany, 1.15338) were used for lipolytic and proteolytic bacterial count analyses. Cultivated Petri dishes were incubated at 30 °C for 48-72 h under aerobic conditions (Halkman and Sagdas, 2011).

#### Sensory analyses

Sensory analysis of the samples was performed with 20 trained panellists using sensory evaluation scorecards. The 9-point hedonic scale was used in the analyses. The scale was established as; 1-3 unacceptable, 4-5 weakly acceptable, 6-7 quite acceptable, 8-9 very good (Tomar, 2018).

#### Statistical evaluation

SPSS 20.0 (SPSS Inc, USA) statistical package program was used to statistically analyze the results. The data obtained from the analyses were evaluated by variance analysis technique in randomized block experimental design. The Duncan test was used to determine the level of difference between groups. The analyses were performed in duplicate and with double parallels.

## Results and discussion

The change in the pH values of yoghurt samples during storage is shown in Figure 1. It was determined that the pH values of yoghurt samples decreased during storage (p<0.05). However, there were no significant differences between the samples (p>0.05). It was found that adding different amounts of plant extracts to yoghurt samples did not have a significant effect on the changes in pH values of the samples during storage (p>0.05). At the beginning of storage, the lowest pH value was 4.29 in 0.3 % and 0.5 % hibiscus extract samples (Figure 1). The lowest pH value (3.95) on the last day of the storage was determined in 0.5 % hibiscus extract sample (Figure 1). Similar to the results obtained in the present study, it was found that 2 %, 4 % and 6 % mint addition in yoghurt production did not significantly affect the pH value of yoghurt (Kumar et al., 2013).

The reason for the decrease in pH values in all samples during the storage period is due to the lactic acid and other organic acids produced by the activities of the starter and non-starter bacteria in yogurt that can ferment lactose (Tomar, 2018).

It was determined that L\* values of the samples (except for control) decreased (p<0.05) and the effect of the addition of plant extract at different ratios on the changes in L\* values of yoghurt samples was statistically significant (Table 1; p<0.05). The increase in the added extract ratio decreased the L\* value further and the maximum decrease was 5.06 % determined in the yoghurt sample containing 0.5 % hibiscus extract.

The addition of 0.1 % mint, basil and hibiscus extracts in yoghurt production did not cause a significant change in the L\* values of the yoghurt samples (p>0.05) (Table 1). However, it was found that 0.3 % and 0.5 % basil and hibiscus extract

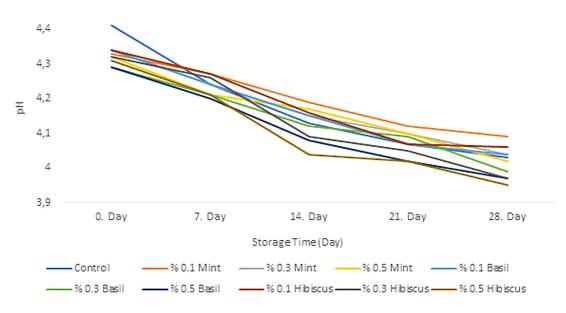


FIGURE 1. Changes in the pH values of samples during storage

Analysis	Sample	Storage period							
		Day O	Day 7	Day 14	Day 21	Day 28			
	Control	90.90±0.10 <sup>aAB</sup>	90.75±0.87ªA	90.99±1.32ªA	90.95±1.39ªA	91.00±1.32ªA			
	0.1 % Mint	90.24±1.09ªAB	89.10±0.54ª <sup>A</sup>	88.89±2.87ªA	87.52±4.09ªA	89.94±2.87ªA			
	0.3 % Mint	88.69±1.70 <sup>abCD</sup>	90.89±0.43ªA	89.24±1.29 <sup>abA</sup>	87.87±0.78 <sup>bA</sup>	87.28±1.29bAB			
	0.5 % Mint	87.05±0.68 <sup>abEF</sup>	90.22±0.10 <sup>aA</sup>	87.25±0.86 <sup>abB</sup>	86.02±3.50 <sup>abB</sup>	87.23±0.86 <sup>bAB</sup>			
alue	0.1 % Basil	90.09±0.67 <sup>aAB</sup>	85.78±1.32 <sup>aCD</sup>	89.84±1.03 <sup>aA</sup>	88.53±3.40 <sup>aA</sup>	89.13±1.03ªA			
L* Value	0.3 % Basil	89.60±0.26 <sup>aC</sup>	86.89±4.50 <sup>aBCD</sup>	85.64±5.48 <sup>aB</sup>	89.34±2.35 <sup>aA</sup>	89.03±5.48 <sup>aAB</sup>			
	0.5 % Basil	89.59±0.89 <sup>aCD</sup>	84.38±0.16 <sup>bD</sup>	85.11±4.75 <sup>ab B</sup>	89.37±0.94 <sup>abA</sup>	86.86±4.75 <sup>abB</sup>			
	0.1 % Hibiscus	91.12±0.40ªA	89.32±1.86ªAB	81.94±10.49ªA	89.46±1.29 <sup>aA</sup>	87.14±10.49ª			
	0.3 % Hibiscus	88.49±0.01 <sup>aDE</sup>	90.75±0.30ªA	87.58±1.58ªA	84.79±6.63 <sup>aB</sup>	87.07±1.58ªAB			
	0.5 % Hibiscus	86.92±0.10 <sup>aF</sup>	88.17±0.47 <sup>aBC</sup>	85.91±0.63 <sup>aB</sup>	85.39±3.44 <sup>aB</sup>	81.86±0.63 <sup>aC</sup>			
	Control	1.74±0.10 <sup>bD</sup>	2.01±0.16 <sup>aD</sup>	1.71±0.12 <sup>bD</sup>	1.64±0.04 <sup>bD</sup>	1.83±0.12 <sup>abD</sup>			
	0.1 % Mint	1.12±0.13 <sup>bE</sup>	0.92±0.20 <sup>bE</sup>	0.97±0.05 <sup>bE</sup>	1.48±0.11 <sup>aDE</sup>	1.66±0.05 <sup>aDE</sup>			
	0.3 % Mint	0.29±0.08 <sup>cG</sup>	0.24±0.04 <sup>cEF</sup>	0.40±0.09 <sup>cF</sup>	0.81±0.11 <sup>bF</sup>	1.15±0.09 <sup>aE</sup>			
	0.5 % Mint	-0.72±0.08 <sup>bH</sup>	-0.45±0.25 <sup>bF</sup>	-0.57±0.05 <sup>bH</sup>	-0.20±0.09 <sup>aG</sup>	0.03±0.05ªF			
alue	0.1 % Basil	1.03±0.12 <sup>bcEF</sup>	1.12±0.16 <sup>bcE</sup>	0.83±0.08 <sup>cE</sup>	1.20±0.16 <sup>bEF</sup>	1.56±0.08 <sup>aDE</sup>			
a* Value	0.3 % Basil	0.82±0.03 <sup>bF</sup>	0.78±2.43 <sup>bE</sup>	0.68±0.23 <sup>bEF</sup>	1.24±0.12 <sup>aE</sup>	1.37±0.23ªE			
	0.5 % Basil	-0.50±0.06 <sup>bH</sup>	-0.24±0.80 <sup>abE</sup>	-0.13±0.34 <sup>abG</sup>	0.14±0.11 <sup>bG</sup>	0.15±0.34 <sup>bF</sup>			
	0.1 % Hibiscus	3.37±0.06 <sup>aC</sup>	3.17±1.44 <sup>bC</sup>	2.89±0.23 <sup>aC</sup>	3.11±0.21 <sup>abC</sup>	3.44±0.23ªC			
	0.3 % Hibiscus	5.04±0.35 <sup>aB</sup>	4.53±0.08 <sup>bB</sup>	4.78±0.10 <sup>abB</sup>	4.79±0.16 <sup>abB</sup>	5.26±0.10 <sup>aB</sup>			
	0.5 % Hibiscus	6.66±0.16 <sup>aA</sup>	6.65±0.36 <sup>aA</sup>	6.62±0.03 <sup>aA</sup>	6.53±0.34ªA	6.68±0.03ªA			
	Control	1.93±0.08ªE	1.47±0.08 <sup>bE</sup>	1.93±0.26 <sup>aD</sup>	1.84±0.21 <sup>abD</sup>	2.22±0.26 <sup>aD</sup>			
	0.1 % Mint	3.35±0.12 <sup>abD</sup>	3.83±1.66 <sup>abC</sup>	4.01±0.16 <sup>aC</sup>	3.17±0.66 <sup>bC</sup>	3.79±0.16 <sup>abC</sup>			
	0.3 % Mint	5.44±0.34 <sup>aC</sup>	5.59±0.09 <sup>aB</sup>	5.75±0.25 <sup>aB</sup>	5.36±0.28 <sup>aB</sup>	5.90±0.25ª <sup>B</sup>			
	0.5 % Mint	8.15±0.06 <sup>bA</sup>	8.03±0.26 <sup>bA</sup>	8.76±0.03 <sup>abA</sup>	8.87±0.61ªA	8.66±0.03 <sup>abA</sup>			
b* Value	0.1 % Basil	3.18±0.23 <sup>aD</sup>	3.15±0.36 <sup>aCD</sup>	3.14±0.60ªC	3.24±0.46 <sup>aC</sup>	3.37±0.60ªC			
> *d	0.3 % Basil	3.19±0.28 <sup>aD</sup>	2.84±0.02 <sup>aB</sup>	3.46±0.66 <sup>aC</sup>	3.03±0.35 <sup>aC</sup>	3.60±0.66 <sup>aC</sup>			
	0.5 % Basil	5.90±0.01 <sup>aB</sup>	5.76±0.76 <sup>aE</sup>	5.68±0.82 <sup>aB</sup>	5.86±0.31 <sup>aB</sup>	6.45±0.82ªB			
	0.1 % Hibiscus	0.82±0.11 <sup>bF</sup>	1.27±0.37 <sup>abE</sup>	1.47±0.60 <sup>abDE</sup>	1.72±0.40 <sup>aDE</sup>	1.23±0.60 <sup>aDE</sup>			
	0.3 % Hibiscus	0.78±0.16 <sup>aF</sup>	0.94±0.73ªE	0.95±0.25 <sup>aDE</sup>	0.81±0.30ªEF	1.01±0.25 <sup>aEF</sup>			
	0.5 % Hibiscus	0.26±0.23 <sup>bG</sup>	0.69±0.40ªE	0.78±0.22ª E	0.68±0.15 <sup>aF</sup>	0.76±0.22ªF			

TABLE 1. L\*, a\* and b\* values of the yoghurt samples during storage time (n=3)

(Mean  $\pm$  standard deviation) Different lower-case letters in the same row and different upper-case letters in the same column indicate statistically significant differences (p<0.05)

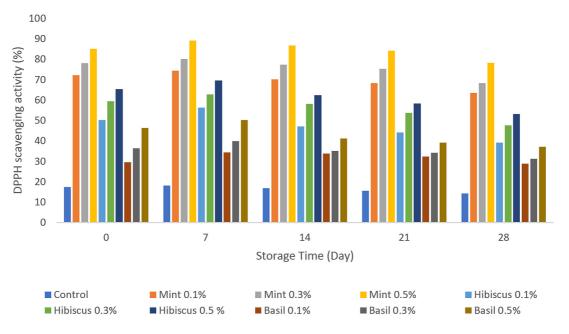


FIGURE 2. Inhibitory percent of DPPH yoghurt samples during storage

significantly decreased the L value of the yoghurt (p<0.05). Similar to the results obtained in the present study, Sisman (2009) stated that the addition of 0.7 % and 1.0 % mint in strained yoghurt decreased the L\* value (Table 1).

The a\* values of all samples increased during storage (Table 1; p<0.05). The highest a\* value in the first and the last days of the storage were 1.74 and 1.83, respectively in the control sample, whereas the lowest a\* values were -0.72 and 0.03, respectively, in the sample containing 0.05 % mint extract. The use of mint and basil extract in yoghurt samples decreased a\* value compared to the control group samples (p<0.05). As the concentrations of mint and basil extracts increased, there was a significant change towards green colour (p<0.05). The addition of hibiscus extract to yoghurt samples increased the redness value significantly (p<0.05). As the concentration of hibiscus extract increased, the colour of yoghurt samples showed significant changes towards red colour (p<0.05). The increase in  $\alpha^*$  was thought to be due to anthocyanin's found in hibiscus.

It was determined that there was an increase in b\* values of yoghurt samples during the storage period (Table 1; p<0.05). The sample with the highest b\* value increase was the sample containing 0.5 % basil extract. This sample was followed by 0.5 % mint and 0.5 % hibiscus extracts with 0.51 and 0.50, respectively. The use of mint and basil extract in yoghurt production decreased b\* value compared to the control group samples significantly (p<0.05). As the concentrations of mint and basil extracts increased, there was a significant change towards yellow colour (p<0.05). Hibiscus extract addition to yoghurt samples decreased the b\* values. As the concentration of hibiscus extract increased, the colour of yoghurt samples showed significant changes towards blue colour (p<0.05). The increase in b\* was thought to be due to chlorophyll found in the extracts.

The reason for the changes in L \*, a \* and b \* values in yogurt samples during storage may be due to the breakdown of colour pigments and polyphenolic components with an increase in organic acid concentration (decrease in pH value) (Watawana et al., 2018).

The change in the antioxidant values of the 2-diphenyl-1-picrihydrazyl (DPPH) radical scavenging capacity of the samples during storage is shown in Figure 2. Yoghurt has been reported to have antioxidant activity due to bioactive peptides formed by fermentation (Yerlikaya et al., 2011).

The antioxidant activity (DPPH) values of yoghurt samples increased in the first seven days of storage and decreased in the following days (Figure 2; p<0.05). It was determined that antioxidant activity values of the samples produced by adding mint extract during storage were higher than those TABLE 2. TAMB, LAB and Lactococcus/Streptococcus counts (log cfu/g) of yoghurt samples during storage

sis	Sample	Storage period						
Analysis		Day 0	Day 7	Day 14	Day 21	Day 28		
TAMB counts	Control	5.45±0.02ªA	5.34±0.02 <sup>aAB</sup>	5.18±0.04 <sup>bAB</sup>	5.15±0.05 <sup>bA</sup>	5.11±0.04 <sup>bA</sup>		
	0.1 % Mint	5.45±0.04ªA	5.38±0.01 <sup>abA</sup>	5.29±0.03 <sup>bA</sup>	5.13±0.03 <sup>cA</sup>	5.09±0.02 <sup>cA</sup>		
	0.3 % Mint	5.45±0.02ªA	5.37±0.01ªA	5.27±0.04 <sup>bA</sup>	5.13±0.01 <sup>cbA</sup>	5.07±0.03 <sup>cA</sup>		
	0.5 % Mint	5.43±0.01ªA	5.25±0.02 <sup>bABC</sup>	5.04±0.02 <sup>cABC</sup>	5.00±0.01 <sup>cAB</sup>	4.93±0.04 <sup>dAB</sup>		
	0.1 % Basil	5.44±0.02ªA	5.32±0.02 <sup>abAB</sup>	5.23±0.06 <sup>bcAB</sup>	5.07±0.06 <sup>cAB</sup>	4.90±0.08 <sup>dAB</sup>		
	0.3 % Basil	5.36±0.06 <sup>aABC</sup>	5.34±0.05 <sup>aAB</sup>	5.19±0.15 <sup>abAB</sup>	4.95±0.02 <sup>bAB</sup>	4.88±0.04 <sup>cAB</sup>		
	0.5 % Basil	5.42±0.07 <sup>aAB</sup>	5.39±0.08ªA	4.99±0.03 <sup>bBC</sup>	4.84±0.07 <sup>bAB</sup>	4.78±0.06 <sup>cB</sup>		
	0.1 % Hibiscus	5.29±0.03 <sup>aBC</sup>	5.22±0.01 <sup>aBC</sup>	4.62±0.09 <sup>bD</sup>	4.48±0.21 <sup>cD</sup>	4.45±0.02 <sup>cD</sup>		
	0.3 % Hibiscus	5.28±0.02 <sup>aC</sup>	5.11±0.01 <sup>bCD</sup>	4.88±0.06°C	4.74±0.02 <sup>cBC</sup>	4.65±0.10 <sup>cCB</sup>		
	0.5 % Hibiscus	5.23±0.02 <sup>aC</sup>	5.02±0.05 <sup>abD</sup>	4.83±0.05 <sup>bCD</sup>	4.47±0.04 <sup>cC</sup>	4.41±0.09℃		
LAB counts	Control	5.11±0.05 <sup>aAB</sup>	5.04±0.03 <sup>abAB</sup>	4.78±0.14 <sup>bA</sup>	4.73±0.05 <sup>bA</sup>	4.70±0.07 <sup>cAB</sup>		
	0.1 % Mint	5.12±0.01 <sup>aAB</sup>	5.06±0.03 <sup>aAB</sup>	4.91±0.05 <sup>bA</sup>	4.78±0.02 <sup>cA</sup>	4.62±0.03 <sup>dB</sup>		
	0.3 % Mint	5.07±0.03 <sup>aAB</sup>	4.98±0.01 <sup>aAB</sup>	4.92±0.02 <sup>bA</sup>	4.77±0.03 <sup>cA</sup>	4.60±0.05 <sup>dAB</sup>		
	0.5 % Mint	5.06±0.04 <sup>aAB</sup>	5.04±0.01 <sup>aAB</sup>	4.84±0.06 <sup>aA</sup>	4.47±0.15 <sup>bA</sup>	4.43±0.05 <sup>bC</sup>		
	0.1 % Basil	5.11±0.05 <sup>aAB</sup>	5.07±0.07 <sup>abAB</sup>	4.93±0.07 <sup>bA</sup>	4.93±0.02 <sup>bA</sup>	4.88±0.06 <sup>cA</sup>		
	0.3 % Basil	5.16±0.06 <sup>aAB</sup>	5.05±0.03 <sup>aA</sup>	4.84±0.03 <sup>bA</sup>	4.67±0.06 <sup>bA</sup>	4.62±0.12 <sup>cB</sup>		
	0.5 % Basil	5.13±0.04 <sup>aAB</sup>	5.05±0.03 <sup>aAB</sup>	4.74±0.26 <sup>bA</sup>	4.67±0.33 <sup>bcA</sup>	4.55±0.07 <sup>cBC</sup>		
	0.1 % Hibiscus	5.09±0.07 <sup>aA</sup>	5.05±0.08 <sup>aAB</sup>	4.75±0.10 <sup>bA</sup>	3.04±0.14 <sup>cBC</sup>	2.98±0.05 <sup>dD</sup>		
	0.3 % Hibiscus	5.01±0.01 <sup>aAB</sup>	4.97±0.06 <sup>aAB</sup>	4.81±0.00 <sup>bA</sup>	3.04±0.02 <sup>cB</sup>	2.85±0.05 <sup>dE</sup>		
	0.5 % Hibiscus	4.99±0.01 <sup>aB</sup>	4.90±0.02 <sup>aB</sup>	4.28±0.14 <sup>bB</sup>	2.53±0.04 <sup>cC</sup>	2.33±0.04 <sup>dF</sup>		
Lactococcus/Streptococcus counts	Control	4.61±0.02ªA	4.35±0.04 <sup>bA</sup>	4.25±0.03 <sup>bcA</sup>	4.16±0.06 <sup>cAB</sup>	4.11±0.08 <sup>dAB</sup>		
	0.1 % Mint	4.53±0.08 <sup>aAB</sup>	4.38±0.05 <sup>abA</sup>	4.28±0.03 <sup>abA</sup>	4.12±0.12 <sup>bAB</sup>	4.06±0.09 <sup>cAB</sup>		
	0.3 % Mint	4.55±0.01 <sup>aAB</sup>	4.45±0.01 <sup>abA</sup>	4.34±0.03 <sup>bA</sup>	4.16±0.06 <sup>cAB</sup>	3.98±0.11 <sup>dAB</sup>		
	0.5 % Mint	4.51±0.01 <sup>aAB</sup>	4.30±0.03 <sup>aAB</sup>	3.63±0.07 <sup>bAB</sup>	3.14±0.09 <sup>cBC</sup>	3.83±0.03 <sup>dAB</sup>		
	0.1 % Basil	4.42±0.06 <sup>aABC</sup>	4.45±0.00ªA	4.40±0.02ªA	4.29±0.05 <sup>aAB</sup>	4.06±0.10 <sup>dAB</sup>		
	0.3 % Basil	4.30±0.05 <sup>aBCD</sup>	4.28±0.04 <sup>aAB</sup>	4.50±0.67ªA	4.76±0.15ªA	3.91±0.08 <sup>cAB</sup>		
	0.5 % Basil	4.40±0.02 <sup>aABCD</sup>	4.08±0.14 <sup>abBC</sup>	3.68±0.28 <sup>bA</sup>	3.49±0.08 <sup>bABC</sup>	3.76±0.06 <sup>dAB</sup>		
	0.1 % Hibiscus	4.34±0.08 <sup>aABCD</sup>	4.23±0.02 <sup>aAB</sup>	3.98±0.05 <sup>bA</sup>	3.71±0.08 <sup>cAB</sup>	3.56±0.05 <sup>dAB</sup>		
Γας	0.3 % Hibiscus	4.14±0.12 <sup>aCD</sup>	3.86±0.09 <sup>abCD</sup>	3.56±0.17 <sup>bAB</sup>	2.15±0.18 <sup>cCD</sup>	2.06±0.09 <sup>dAB</sup>		
	0.5 % Hibiscus	4.12±0.15 <sup>aD</sup>	3.70±0.02 <sup>bD</sup>	2.69±0.08 <sup>cbB</sup>	0.78±1.11 <sup>dD</sup>	0.16±0.02 <sup>eAB</sup>		

TAMB: Total aerobic mesophilic bacteria (Mean  $\pm$  standard deviation), LAB: Lactic acid bacteria, (Mean  $\pm$  standard deviation) Different lower-case letters in the same row and different upper-case letters in the same column indicate statistically significant differences (p<0.05)

of the other samples and the antioxidant activity value increased with the increasing ratio of mint extract (p<0.05). Similarly, Amirdivani and Baba (2011) reported that yogurt samples with plant extract added had higher antioxidant activity. The researchers stated that the presence of higher antioxidant activity in the samples containing plant

extract may depend on the phytochemical content and microbial metabolic activity of the plants. On the 0<sup>th</sup> and 28<sup>th</sup> days of the storage, it was determined that the 0.5 % mint extract samples had the highest antioxidant activity with 85.13 % and 78.19%, respectively. Ismail et al. (2004) reported that they detected high amounts of  $\alpha$ -tocopherol, TABLE 3. Proteolytic and lipolytic bacteria counts of yoghurt samples during storage (log cfu/g)

is.	Sample	Storage period							
Analysis		Day 0	Day 7	Day 14	Day 21	Day 28			
Proteolytic bacteria counts	Control	3.72±0.17ªA	3.45±0.21 <sup>abA</sup>	2.92±0.11 <sup>bcA</sup>	2.69±0.12 <sup>cA</sup>	2.36±0.12 <sup>dA</sup>			
	0.1 % Mint	2.74±0.06 <sup>aB</sup>	2.69±0.01 <sup>aBC</sup>	2.29±0.02 <sup>bA</sup>	0.00±0.00°C	0.00±0.00°C			
	0.3 % Mint	2.77±0.03ªB	2.26±0.08 <sup>bCD</sup>	1.60±0.03 <sup>cAB</sup>	0.00±0.00 <sup>dC</sup>	0.00±0.00 <sup>dC</sup>			
	0.5 % Mint	2.66±0.03ªB	1.56±0.08 <sup>bE</sup>	0.00±0.00°C	0.00±0.00℃	0.00±0.00°C			
	0.1 % Basil	2.85±0.08 <sup>aB</sup>	2.89±0.07 <sup>aB</sup>	2.69±0.04ªA	1.53±0.07 <sup>bB</sup>	1.31±0.06 <sup>cB</sup>			
	0.3 % Basil	2.78±0.03ªB	2.81±0.03ªB	2.06±0.12 <sup>bAB</sup>	1.75±0.18 <sup>bB</sup>	1.68±0.12 <sup>cB</sup>			
	0.5 % Basil	2.86±0.11ªB	2.56±0.26ªBC	0.81±1.14 <sup>abBC</sup>	0.00±0.00 <sup>bC</sup>	0.00±0.00°C			
	0.1 % Hibiscus	2.76±0.04 <sup>aB</sup>	1.95±0.19 <sup>bDE</sup>	0.00±0.00°C	0.00±0.00℃	0.00±0.00 <sup>dC</sup>			
	0.3 % Hibiscus	2.60±0.09 <sup>aBC</sup>	1.49±0.02 <sup>bE</sup>	0.00±0.00°C	0.00±0.00℃	0.00±0.00°C			
	0.5 % Hibiscus	2.23±0.16 <sup>aC</sup>	0.00±0.00 <sup>bF</sup>	0.00±0.00 <sup>bC</sup>	0.00±0.00 <sup>bC</sup>	0.00±0.00°C			
Lipolytic bacteria counts	Control	4.06±0.03ªA	4.00±0.00ªA	3.97±0.10 <sup>abA</sup>	3.78±0.25 <sup>bA</sup>	3.43±0.15 <sup>bB</sup>			
	0.1 % Mint	3.48±0.19 <sup>aCD</sup>	3.45±0.08 <sup>aC</sup>	3.20±0.08 <sup>abA</sup>	2.83±0.05 <sup>bB</sup>	2.57±0.08 <sup>cB</sup>			
	0.3 % Mint	3.59±0.05 <sup>aBCD</sup>	3.39±0.06 <sup>abC</sup>	3.15±0.15 <sup>bA</sup>	3.04±0.06 <sup>bB</sup>	2.91±0.05 <sup>cB</sup>			
	0.5 % Mint	3.49±0.09 <sup>aCD</sup>	2.93±0.15 <sup>bD</sup>	1.73±0.08 <sup>cB</sup>	0.00±0.00 <sup>dD</sup>	0.00±0.00 <sup>dD</sup>			
	0.1 % Basil	3.50±0.20 <sup>aCD</sup>	3.54±0.04ªBC	3.41±0.05ªA	3.21±0.04 <sup>aAB</sup>	3.01±0.14 <sup>bAB</sup>			
	0.3 % Basil	3.76±0.05ªABC	3.68±0.10 <sup>abBC</sup>	3.61±0.13 <sup>abA</sup>	2.94±0.12 <sup>bB</sup>	2.76±0.09 <sup>bAB</sup>			
	0.5 % Basil	3.92±0.09ªAB	3.80±0.01 <sup>abAB</sup>	3.15±0.24 <sup>bA</sup>	1.67±0.18 <sup>cBC</sup>	1.32±0.08 <sup>dBC</sup>			
	0.1 % Hibiscus	3.38±0.10 <sup>aCD</sup>	2.13±0.05 <sup>abE</sup>	1.59±0.05 <sup>abB</sup>	0.74±1.04 <sup>bCD</sup>	0.00±0.00 <sup>bD</sup>			
	0.3 % Hibiscus	3.25±0.01ªD	1.84±0.03 <sup>abEF</sup>	0.74±1.04 <sup>bBC</sup>	0.00±0.00 <sup>bD</sup>	0.00±0.00 <sup>cD</sup>			
	0.5 % Hibiscus	3.21±0.02 <sup>aD</sup>	1.60±0.11 <sup>bF</sup>	0.00±0.00°C	0.00±0.00 <sup>cD</sup>	0.00±0.00 <sup>cD</sup>			

(Mean  $\pm$  standard deviation) Different lower-case letters in the same row and different upper-case letters in the same column indicate statistically significant differences (p<0.05)

 $\beta$ -carotene and ferulic acid with antioxidant activity in peppermint composition.

After the 7<sup>th</sup> day of storage, the antioxidant activity of the samples decreased. Yildiz and Eyduran (2009) attributed this decrease in antioxidant activity to degradation in phenolic compounds with antioxidant activity, while Yüksel et al. (2010) attributed an increase in protein-polyphenol interaction. Depending on this situation, it is suggested that it will be appropriate to consume these yogurts within seven days in order to health benefit from yoghurt with herbal extract.

Total aerobic mesophilic bacteria (TAMB) counts of all samples decreased during storage (Table 2; p<0.05). Hibiscus extract was found to be the most

effective on the TAMB counts in all the plant extracts used in the yoghurt production. The increase in the extract ratio added to the yoghurt showed a significant effect on the decrease in the TAMB counts (p<0.05).

Immediately after yoghurt production, the TAMB counts did not change significantly with the addition of plant extracts in yoghurt (p>0.05). At the end of storage, samples containing 0.5 % basil and 0.1 %, 0.3 % and 0.5 % hibiscus were found to have significantly lower TAMB counts compared to those of the other samples (p<0.05). At the end of the storage period, the lowest TAMB counts was 4.41 log cfu/g determined in the samples added g 0.5 % hibiscus extract whereas the highest TAMB counts

were determined in the control sample with  $5.11 \log cfu/g$ .

Lactic acid bacteria count of all yoghurt samples decreased during storage (Table 2; p<0.05). Compared to other plant extracts, the lactic acid bacteria count of the samples containing hibiscus extract were significantly lower (p<0.05). In addition, the number of lactic acid bacteria decreased as the ratio of extract increased (p<0.05). At the beginning and at the end of storage, the lowest lactic acid bacteria counts were found in the samples containing 0.5 % hibiscus extract with 4.99 and 2.33 log cfu/g, respectively.

During the 28-day storage period, Lactococcus/ Streptococcus counts decreased (Table 2; p<0.05). As the ratio of plant extract added to the samples increased, Lactococcus/Streptococcus counts decreased (p<0.05). Lactococcus/Streptococcus counts were found to be significantly lower in samples containing 0.5 % mint, 0.5 % basil, 0.3 % and 0.5 % hibiscus extracts at the end of storage period (p<0.05). The highest decrease during the storage period was found in the yoghurt samples produced by adding Hibiscus extract (p<0.05). Compared to mint and basil extracts, hibiscus extract was more effective in decreasing the Lactococcus/Streptococcus counts. At the end of storage, the lowest Lactococcus / Streptococcus bacteria count was 0.16 log cfu/g, determined in 0.5 % hibiscus extract. Simsek et al. (2007) have reported Lactobacillus/ Streptococcus counts at the end of storage in yoghurt samples containing 0.5 % and 0.2 % mint as 4-5 log cfu/g.

Proteolytic bacteria count of yoghurt samples decreased during storage (Table 3; p<0.05). At the end of the storage period, the highest proteolytic bacteria count was found to be 2.36 log cfu/g, in the control sample. Among the three different plant extracts used in the production of yoghurt, it was determined that the extract which halted the proteolytic bacterial growth in the fastest way was hibiscus extract and mint and basil followed hibiscus respectively.

Especially, the increase in concentration was found to inhibit proteolytic bacteria growth at a higher level (p<0.05). Proteolytic bacteria were not detected after 7 days of storage in samples containing 0.5 % hibiscus extract, while proteolytic bacteria were not detected after 14 days of storage in samples containing 0.1 % and 0.3 % hibiscus and 0.5 % mint extracts.

The count of lipolytic bacteria decreased in all samples during storage (Table 3; p<0.05). However, lipolytic bacteria were less affected by plant extracts immediately after production compared to proteolytic bacteria. Yoghurt samples containing mint, basil, hibiscus extracts were found to contain significantly lower lipolytic bacteria counts during storage compared to those of the control group samples (p<0.05). The lowest decrease was detected in the control sample during the 28-day storage whereas the highest decrease was found in the yoghurt sample containing 0.5 % hibiscus extract. Lipolytic bacteria could not be detected after 14 days of storage in yoghurt samples containing 0.5% hibiscus extract, and after 21 days of storage in yoghurt samples containing 0.3 % hibiscus and 0.5 % mint. Compared to peppermint and basil extracts, hibiscus extract had a higher inhibitory effect on lipolytic bacteria. On the 28<sup>th</sup> day of the storage, it was determined that lipolytic bacteria counts decreased to 0 log cfu/g in all samples containing hibiscus extract (p<0.05). The effect of hibiscus extract on the lipolytic bacteria was followed by mint and basil extract was not as effective as the other two extracts even at the highest concentration. In addition, the effect of the increase in the ratio of the extract on lipolytic bacteria was found to be more effective in a shorter time (p < 0.05).

The detection of a lower number of microorganisms in samples containing Hibiscus may have been due to the antimicrobial effective compounds in the hibiscus. The presence and quantities of alkaloids, phenolic compounds, flavonoids, cyanidins and saponins found in *Hibiscus surattensis* L. was demonstrated with previously performed studies (Abdallah, 2016). Akarca (2019) reported that hibiscus has antibacterial activity due to  $\beta$ -caryophyllene, menthol, methyl salicylate, camphor and germacrene and hibiscus essential oil, had higher antibacterial effect against many foodborne pathogens. Patel et al. (2012) reported that Hibiscus extracts are effective against various pathogens.

Yoghurt can easily be contaminated with microorganisms from equipment used in different stages of production, packaging material and air. These contaminating microorganisms proliferate during storage and adversely affect product quality and

		Samples										
	Control	0.1 % Mint	0.3 % Mint	0.5 % Mint	0.1 % Basil	0.3 % Basil	0.5 % Basil	0.1 % Hibiscus	0.3 % Hibiscus	0.5 % Hibiscus		
					Appear	ance						
0	8.82±0.16ª	8.73±0.08 <sup>a</sup>	7.02±0.26ª	5.27±0.48ª	8.31±0.16ª	6.86±0.31ª	4.77±0.14ª	8.78±0.18 <sup>a</sup>	7.27±0.17ª	6.13±0.19ª		
7	8.76±0.43ª	8.51±0.13ª	6.62±0.09 <sup>b</sup>	4.38±0.29 <sup>b</sup>	8.16±0.25 <sup>ab</sup>	6.31±0.11 <sup>b</sup>	3.68±0.18 <sup>b</sup>	8.64±0.21 <sup>ab</sup>	7.02±0.15 <sup>b</sup>	5.42±0.14 <sup>b</sup>		
14	8.12±0.12 <sup>b</sup>	7.92±0.16 <sup>b</sup>	5.74±0.22°	3.86±0.23℃	7.71±0.19 <sup>b</sup>	5.54±0.12℃	3.13±0.33°	8.12±0.07 <sup>b</sup>	6.59±0.12°	5.01±0.08°		
21	7.80±0.28℃	7.33±0.13°	5.52±0.14 <sup>cd</sup>	3.38±0.23 <sup>cd</sup>	7.16±0.43°	5.31±0.22 <sup>cd</sup>	2.98±0.45 <sup>cd</sup>	7.74±0.11 <sup>bc</sup>	6.50±0.24°	4.52±0.11 <sup>cd</sup>		
28	6.92±0.13 <sup>d</sup>	6.65±0.10 <sup>d</sup>	5.32±0.23 <sup>d</sup>	3.22±0.36d	6.41±0.13 <sup>d</sup>	5.02±0.13 <sup>d</sup>	2.82±0.31d	7.05±0.16°	5.48±0.13 <sup>d</sup>	4.18±0.32 <sup>d</sup>		
Taste and odor												
0	8.02±0.55ª	8.32±0.33ª	7.02±0.27 <sup>b</sup>	5.52±0.32 <sup>b</sup>	7.73±0.21 <sup>ab</sup>	6.41±0.32ª	4.46±0.35ª	8.55±0.27ª	7.13±0.36ª	4.06±0.15ª		
7	7.94±0.36 <sup>ab</sup>	8.16±0.26 <sup>ab</sup>	7.11±0.16ª	5.54±0.12ª	7.81±0.17ª	6.50±0.13ª	4.50±0.34ª	8.62±0.21ª	7.18±0.19ª	4.11±0.04ª		
14	7.66±0.41 <sup>b</sup>	8.02±0.38 <sup>b</sup>	5.75±0.28°	4.15±0.17℃	7.55±0.35⁵	5.05±0.22⁵	3.25±0.12 <sup>b</sup>	8.11±0.33 <sup>b</sup>	6.61±0.25 <sup>b</sup>	3.62±0.22 <sup>b</sup>		
21	6.91±0.66°	7.71±0.38℃	5.13±0.41 <sup>cd</sup>	3.82±0.34 <sup>cd</sup>	7.23±0.28℃	4.86±0.37 <sup>bc</sup>	3.08±0.25⁵	7.84±0.18°	6.03±0.20 <sup>c</sup>	3.14±0.32℃		
28	6.34±0.26 <sup>d</sup>	6.84±0.33 <sup>d</sup>	4.64±0.31 <sup>d</sup>	3.32±0.44 <sup>d</sup>	6.81±0.21 <sup>d</sup>	4.12±0.14 <sup>c</sup>	2.74±0.24 <sup>c</sup>	7.26±0.23 <sup>d</sup>	5.42±0.11 <sup>d</sup>	2.61±0.44 <sup>d</sup>		
		1	1		Textu	ire	1	1				
0	8.42±0.22ª	8.13±0.06ª	7.42±0.13ª	7.04±0.10ª	7.83±0.11ª	7.15±0.06ª	6.71±0.14ª	8.33±0.16 <sup>ab</sup>	7.92±0.07 <sup>ab</sup>	7.36±0.11 <sup>b</sup>		
7	8.46±0.03ª	8.22±0.18ª	7.47±0.11 <sup>b</sup>	7.06±0.09 <sup>b</sup>	7.92±0.14ª	7.17±0.15ª	6.76±0.16 <sup>a</sup>	8.42±0.12ª	8.04±0.06ª	7.64±0.24 <sup>ba</sup>		
14	8.24±0.22 <sup>b</sup>	8.02±0.06 <sup>b</sup>	6.36±0.12°	5.84±0.13 <sup>b</sup>	7.58±0.14 <sup>b</sup>	6.08±0.13 <sup>b</sup>	5.12±0.06 <sup>b</sup>	8.16±0.10 <sup>b</sup>	7.53±0.05 <sup>b</sup>	6.64±0.16°		
21	7.40±0.08℃	7.27±0.11°	6.21±0.08 <sup>cd</sup>	5.28±0.03°	7.04±0.10°	5.62±0.05°	4.68±0.013°	7.77±0.21°	6.92±0.12 <sup>c</sup>	5.88±0.08 <sup>d</sup>		
28	7.04±0.32 <sup>d</sup>	6.74±0.12 <sup>d</sup>	5.76±0.07 <sup>d</sup>	4.91±0.06 <sup>d</sup>	6.34±0.08 <sup>d</sup>	5.04±0.09 <sup>d</sup>	4.11±0.06 <sup>d</sup>	7.14±0.16 <sup>d</sup>	6.16±0.12 <sup>d</sup>	5.21±0.16 <sup>e</sup>		
General appreciation												
0	8.54±0.14ª	8.36±0.05ª	6.15±0.06ª	5.33±0.18ª	8.27±0.10ª	7.16±0.11ª	5.17±0.08ª	8.48±0.15ª	6.36±0.09ª	5.54±0.05ª		
7	8.61±0.21ª	8.42±0.12ª	6.15±0.05ª	4.56±0.19⁵	8.29±0.15ª	7.21±0.08ª	5.17±0.12ª	8.53±0.14ª	6.35±0.13ª	5.56±0.13ª		
14	8.03±0.09 <sup>b</sup>	7.96±0.06 <sup>b</sup>	5.54±0.04 <sup>b</sup>	4.06±0.13℃	7.65±0.09 <sup>b</sup>	5.16±0.12⁵	4.23±0.13 <sup>b</sup>	8.16±0.08 <sup>b</sup>	6.11±0.08 <sup>b</sup>	4.27±0.09 <sup>b</sup>		
21	7.52±0.07℃	7.41±0.03℃	4.61±0.10°	3.64±0.09℃	7.32±0.13℃	4.27±0.07°	3.91±0.05℃	8.01±0.14°	5.31±0.18°	3.79±0.014°		
28	6.82±0.06 <sup>d</sup>	6.54±0.12 <sup>d</sup>	4.42±0.014 <sup>d</sup>	3.52±0.06°	6.11±0.08 <sup>d</sup>	4.02±0.11 <sup>d</sup>	3.12±0.06 <sup>d</sup>	7.04±0.06 <sup>d</sup>	4.76±0.013 <sup>d</sup>	3.61±0.07 <sup>d</sup>		
	1	I	1	1	1				1	1		

(Mean  $\pm$  standard deviation) Different lower-case letters in the same row and different upper-case letters in the same column indicate statistically significant differences (p<0.05)

reduce shelf life. The main bacteria that affect the microbiological quality of the product are lipolytic and proteolytic bacteria. These bacteria cause rancid deterioration as a result of lipolytic activity and yeasty bitter flavor and gas formation as a result of the proteolytic activity causing quality losses in yoghurt (Harasawa et al., 1998; Gurakan, 2010).

Hibiscus extract significantly reduced the development of TAMB, LAB and *Lactococcus/Streptococcus* bacteria, hibiscus was followed by mint and basil extracts. Lipolytic and proteolytic bacterial growth was significantly reduced by the addition of plant extracts. The most effective plant extract against these bacteria was hibiscus, followed by mint whereas the least effective extract was basil extract.

The results of sensory analysis of yoghurt samples by the panellists during the 28 day storage are given in Table 4. Appearance scores increased in the first seven days of storage however decreased in the following days (p<0.05). The sample with the highest appearance scores at the beginning and the end of storage was the control sample. The samples with the highest appearance scores following the control sample were those containing 0.1 % hibiscus, 0.1 % mint and 0.1 % basil extracts, respectively. Appearance scores decreased with the increasing ratios of extract added to the samples (p<0.05).

Although taste and aroma scores increased in the first 7 days of storage, they decreased in the following days (p<0.05). The highest taste and aroma points at the beginning and the end of storage were determined in the samples containing 0.1 % hibiscus extract with 8.55 and 7.26, respectively. This was followed by the samples containing 0.1 % mint extract with 8.32 and 6.84, respectively.

Unlike the other two extracts, yoghurt samples containing 0.1 % basil extract received lower taste and aroma scores. In addition, the increase in the ratio of extract had a negative effect on taste and aroma scores (p<0.05). The samples with the lowest taste and aroma scores were the samples containing 0.5 % extract.

The sensory scores increased in the first 7 days of the storage however decreased in the following days (p<0.05). On the first and last day of storage, the highest texture score was obtained from the control sample with 8.42 and 7.04, respectively. The lowest texture score was obtained from the sample containing 0.5 % extract with 6.71 and 4.11, respectively. The control sample was followed by 0.1 % hibiscus, 0.1 % mint and 0.1 % basil samples (p<0.05). It was determined that texture scores decreased with increasing ratio of extract (p<0.05).

General evaluation scores of yoghurt samples increased in the first 7 days of storage however they decreased in the following days (p<0.05). The lowest decrease during storage was 1.44, determined in 0.1 % hibiscus whereas the highest decrease was 0.3 % determined in basil extract. The control samples received the highest sensory scores, followed by the samples containing 0.1 % hibiscus and 0.1 % mint extracts, respectively. On the other hand, the samples with the lowest sensory score were the samples containing 0.5 % basil extract. As in other sensory scores, the increase in the ratio of extract decreased the general evaluation scores (p<0.05).

## Conclusion

In this study-it was found that the addition of plant extracts increased the antioxidant activity of yoghurt samples. Higher antioxidant activity was measured in samples with mint extract. Plant extracts partially affected the colour values of yoghurts. Additionally, plant extracts affected the microbial counts of the samples. Especially in samples containing basil and hibiscus, lower bacteria counts were found.

The most important point in the addition of herbal extracts to yoghurt samples was their effects on the sensory properties of the samples. According to the results of sensory analysis, the addition of 0.1 % plant extracts to voghurts rated higher scores than that of the control sample, but the addition of 0.3 % and 0.5 % extracts negatively affected the sensory properties. It can be concluded that adding plant extracts had a positive effect on the sensory and functional properties of yoghurt. Therefore, the addition of mint, hibiscus or basil extracts to yoghurt is recommended, as this has the potential to be further developed for consumers as a functional yoghurt with antioxidant properties. Yoghurt producers may use different plants to increase the sensory and functional properties of yoghurt with reference to this study. Thus, it is possible to produce high quality products with increased consumer appreciation. In addition, increasing the antioxidant activity of yoghurt will further enhance the current positive effects of yoghurt on health.

## Utjecaj dodatka biljnih ekstrakata na antioksidacijski potencijal, mikrobiološka i senzorska svojstva tekućeg jogurta

#### Sažetak

Cilj ovog istraživanja bio je ispitati mogućnost dodatka različitih biljnih ekstakata radi poboljšanja funkcionalnih svojstava jogurta. Tekućem jogurtu dodavani su etanolni ekstrakti triju različitih biljnih vrsta (Mentha piperita L., Ocimum basilicum L. i Hibiscus sabdariffa L.) u različitim omjerima (0,1 %, 0,3 % i 0,5 %). Svim uzorcima jogurta određivani su pH vrijednost, parametri boje, antioksidacijski potencjal, mikrobiološka i senzorska svojstva tijekom perioda čuvanja (0, 7, 14, 21 i 28 dan). Najniže pH vrijednosti utvrđene su na početku (4,29) i na kraju (3,95) skladištenja u uzorcima koji sadrže 0,5 % ekstrakta hibiskusa. Dodavanje biljnih ekstrakata u količini 0,3 % i više smanjilo je vrijednost svjetline (L \*) jogurta (p<0,05), a maksimalno smanjenje zabilježeno je u uzorcima jogurta koji sadrže 0,5 % etanolnog ekstrakta hibiskusa. Veće koncentracije dodanih ekstrakta metvice i bosiljka povećavale su vrijednost žutine (b \*) (p<0,05), a dodavanje ekstrakta hibiskusa značajno je povećalo vrijednost crvenila (p <0,05). Etanolni ekstrakt hibiskusa značajno je smanjio rast ukupnih aerobnih mezofilnih bakterija (TAMB), bakterija mliječne kiseline (LAB) te laktokoka i streptokoka (p<0,05). Antioksidativni potencijal svih uzoraka povećala se dodatkom biljnog ekstrakta. Prema rezultatima senzorske analize, dodavanje 0,1 % biljnih ekstrakata jogurtima ocijenjeno je višim rezultatima od kontrolnog uzorka, ali dodavanje 0,3 % i 0,5 % ekstrakta negativno je utjecalo na senzorska svojstva. Može se zaključiti da je dodavanje biljnih ekstrakata pozitivno utjecalo na senzorska i funkcionalna svojstva jogurta.

#### Ključne riječi: jogurt; biljni ekstrakt; antioksidativni potencijal; boja

## References

- Abdallah, E.M. (2016): Antibacterial efficiency of the Sudanese roselle (*Hibiscus sabdariffa* L.), a famous beverage from Sudanese folk medicine. *Journal of Intercultural Ethnopharmacology* 5 (2), 186-190. https://doi.org/10.5455/jice.20160320022623.
- Adegoke, G.O., Musenbi, S., Wachira, S., Muigai, C., Muigai, F., Ngumba, E., Karanja, P., Onyango, C., Adoko, G.M. (2013): Production of probiotic yoghurt flavored with the spice, Aframomum danielli, strawberry and vanilla. Food and Public Health 3 (2), 92-96.
- Akarca, G. (2019). Composition and antibacterial effect on food borne pathogens of *Hibiscus surrattensis* L. calyces essential oil. *Industrial Crops and Products* 137, 285-289.

https://doi.org/10.1016/j.indcrop.2019.05.043

 Akarca, G., Tomar, O., Güney, İ., Erdur, S., Gök, V. (2019): Determination of sensitivity of some food pathogens to spice extracts. *Journal of Food Science and Technology* 56 (12), 5253-5261. https://doi.org/10.1007/s13197-019-03994-1

- Amirdivani, S., Baba, A.S. (2011): Changes in yoghurt fermentation characteristics, and antioxidant potential and in vitro inhibition of angiotensin-1 converting enzyme upon the inclusion of peppermint, dill and basil. *LWT-Food Science and Technology* 44, 1458-1464. http://doi.org/10.1016/j.lwt.2011.01.019
- Apostolidis, E., Kwon, Y.I., Shetty, K. (2007): Inhibitory potential of herb, fruit and fungal- enriched cheese against key enzymes linked to type 2 diabetes and hypertension. *Innovative Food Science and Emerging Technologies* 8, 46-54. http://doi.org/10.1016/j.lfset.2006.06.001
- Cappato, L.P., Ferreira, M.V.S., Pires, R.P., Cavalcanti, R.N., Bisaggio, R.C., Freitas, M.Q., Silva, M.C., Cruz, A.G. (2018): Whey acerola-flavoured drink submitted ohmic heating processing: Is there an optimal combination of the operational parameters? *Food Chemistry* 245, 22-28.
- Chandan, R.C., O'Rell, K. (2013): Ingredients for yoghurt manufacture. In: Chandan, R.C., Kilara, A., editors. Manufacturing yoghurt and fermented milks, John Wiley & Sons pp. 217-237.
- Corroler, D., Mangin, I., Desmasure, N., Gueguen, M. (1998): An ecological study of lactococci isolated from raw milk in the Camembert cheese registered designation of origin area. *Applied and Environmental Microbiology* 64 (12), 4729-4735.

- Freitas, M. (2017): The benefits of yoghurt, cultures, and fermentation (Chapter 24). In: Floch, M.H., Ringel, Y., Walker, W.A., editors. The microbiota in gastrointestinal pathophysiology, Academic Press. pp. 209-223.
- Gao, H.X., Yu, Z.L., He, Q., Tang, S.H., Zeng, W.C. (2018): A potentially functional yoghurt co-fermentation with Gnaphalium affine. *LWT-Food Science and Technology* 91, 423-430.

https://doi.org/10.1016/j.lwt.2018.01.085

- Gupta, A.K., Mishra, R., Singh, A.K., Srivastava, A., Lal, R.K. (2017): Genetic variability and correlations of essential oil yield with agro-economic traits in *Mentha species* and identification of promising cultivars. *Industrial Crops and Products* 95, 71-96. https://doi.org/10.1016/j.indcrop.2016.11.041
- Gurakan, G.C., Altay, N. (2010): Yoghurt microbiology and biochemistry. In Yildiz, F., editor. Development and manufacture of yoghurt and other functional dairy product, CRC Press, Taylor & Francis Group pp. 98-116.
- Gurkan, H., Hayaloglu, A.A. (2017): Volatiles and sensory characteristics of yoghurt manufactured by incorporating basil (Ocimum basilicum L.). International journal of food properties, 20(1), 779-789. https://doi.org/10.1080/10942912.2017.1311344
- Halkman, K., Sağdaz Ö.E. (2011): Food microbiology applications. Prosigma Printing and Promotional Services, Ankara.
- Harasawa, N., Tateba, H., Ishizuka, N., Wakayama, T., Kishino, K., Ono, M. (1998): Flavor deterioration in yoghurt. In: Shahidi, F., Ho, C.T., Chuyen, N.V., editors. Processinduced chemical changes in food, Springer-Verlag Inc. pp. 285-296.
- Halal, A., Tagliazucchi, D. (2018): Impact of in-vitro gastro-pancreatic digestion on polyphenols and cinnamaldehydebioaccessibility and antioxidant activity in stirred cinnamon-fortified yoghurt. LWT - Food Science and Technology (Lebensmittel-Wissenschaft -Technol.), 89, 164-170.
- Hervert-Hernández, D., Goni, I. (2012): Contribution of beverages to the intake of polyphenols and antioxidant capacity in obese women from rural Mexico. *Public Health Nutrition* 15, 6-12. http://dx.doi.org /10.1017/S1368980011001753
- Illupapalayam, V.V., Smith, S.C., Gamlath, S. (2014): Consumer acceptability and antioxidant potential of probiotic-yoghurt with spices. *LWT-Food Science and Technology* 55 (1), 255-262. https://doi.org/10.1016/j.lwt.2013.09.025
- Ismail, A., Marjam, Z. M., Foong, C. W. (2004): Total antioxidant activity and phenolic content in selected vegetables. *Food Chemistry* 87, 581-586.
- ISO (1999): International Standard Organization, 6888-1 Horizontal Method for the Enumeration of Coagulasepositive Staphylococci Technique using Baird Parker Agar Medium, Geneva, Switzerland.
- ISO (2013a): International Standard Organization.
   4833-2:2013 Horizontal method for the enumeration of microorganisms. Part 2: Colony count at 30 degrees C by the surface plating technique. Geneva, Switzerland.

- 23. ISO (2013b): International Standard Organization. 4833-1:2013 Microbiology of the food chain. Horizontal method for the enumeration of microorganisms. Part 1: Colony count at 30 degrees C by the pour plate technique. Geneva, Switzerland.
- Jung, J., Paik, H.D., Yoon, H.J., Jang, H.J., Jeewanthi, R.K.C., Jee, H.S., Li , X., Lee, N.K., Lee, S.K. (2016): Physicochemical characteristics and antioxidant capacity in yoghurt fortified with red ginseng extract. *Korean Journal for Food Science of Animal Resources* 36(3), 412-420.
- https://doi.org/10.5851/kosfa.2016.36.3.412.
  25. Kim, S. H., Lim, H. W., Chon, J. W., Song, K. Y., Seo, K. H. (2019): Sensory Profiles of Dairy Products Supplemented with *Hibiscus sabdariffa* Linnaeus (Roselle) Powder: A Preliminary Study. *Journal of Milk Science and Biotechnology*, 37(4), 247-255. https://doi.org/10.22424/jmsb.2019.37.4.247
- Kneifel, W., Berger, E. (1994): Microbiological criteria of random samples of spices and herbs retailed on the Austrian market. *Journal of Food Protection* 57 (10), 893-901.

http://dx.doi.org /10.4315/0362-028X-57.10.893

- Kumar, T.S., Arvindakshan, P., Sangeetha, A., Pagote, C.N., Rao, K.J. (2013): Development of mint flavoured yoğurt spread. Asian Journal of Dairy and Food Research 32 (1), 19-24.
- Paszczyk, B., Łuczyńska, J., Polak-Śliwińska, M. (2020): The effect of storage on the yoghurt fatty acid profile. *Mljekarstvo* 70 (1), 59-70. https://doi.org/10.15567/mljekarstvo.2020.0106
- Patel, R., Patel, A., Vaghasiya, D., Nagee, A. (2012): Antimicrobial evaluation of hibiscus rosa-sinensis plant extracts against some pathogenic bacteria. *Bulletin of Environmental and Scientific Research* 1(3-4), 14-17.
- 30. Senadeera, S.S., Prasanna, P.H.P., Jayawardana, N.W.I.A., Gunasekara, D.C.S., Senadeera, P., Chandrasekara, A. (2018): Antioxidant, physicochemical, microbiological, and sensory properties of probiotic yoghurt incorporated with various Annona species pulp. Heliyon 4 (11), e00955. https://doi.org/10.1016/j.heliyon.2018.e00955
- Senel, E., Gürsel, A., Yaman, Ş., Tamuçay, B. (2006): Effect of using a biopreservative culture on some properties of set-type yoghurt. *The Journal of Food* 31 (1), 21-26.
- Simsek, B., Sagdic, O., Ozcelik, S. (2007): Survival of Escherichia coli O157:H7 during the storage of Ayran produced with different spices. *Journal of Food Engineering* 78 (2), 676-680. https://doi.org/10.1016/j.jfoodeng.2005.11.005
- Sindi, H.A., Marshall, L.J., Michael, R.A., Morgan, M.R.A. (2014): Comparative chemical and biochemical analysis of extracts of *Hibiscus sabdariffa*. *Food Chemistry* 164 (1), 23-29.

https://doi.org/10.1016/j.foodchem.2014.04.097

 Singh, G., Kapoor, I.P.S., Singh, P. (2011): Effect of volatile oil and oleoresin of anise on the shelf life of yoghurt. *Journal of Food Processing and Preservation* 35 (6), 778-783.

- 35. Sisman, I.E. (2009): A research about spreadable aromatized concentrated (torba) yoghurt production. M.Sc. Thesis, Selcuk University, Graduate School of Natural and Applied Sciences, Konya, Turkey.
- Soilhi, Z., Rhimi, A., Heuskin, S., Fauconnier, M.L., Mekkia, M. (2019): Essential oil chemical diversity of Tunisian Mentha spp. collection. Industrial Crops and Products 131, 330-340.

https://doi.org/10.1016/j.indcrop.2019.01.041

- Tapsell, L., Hemphill, I., Cobiac, L., Patch, C., Sullivan, D., Fenech, M. (2006): Health benefits of herbs and spices: the past, the present, the future. *Medical Journal of Australia* 185 (4), 4-24.
- Tomar, O. (2018): The effects of probiotic cultures on the organic acid content, texture profile and sensory attributes of Tulum cheese. *International Journal of Dairy Technology* 70 (2), 218-228. https://doi.org/10.1111/1471-0307.12574
- 39. Varga, F., Carović-Stankoa, K., Ristićc, M., Grdišaa, M., Liberb, Z., Šatovića Z. (2017): Morphological and biochemical intraspecific characterization of *Ocimum basilicum* L. *Industrial Crops and Products* 109, 611-618. https://doi.org/10.1016/j.indcrop.2017.09.018
- 40. Vieira, P., Pinto, C.A., Lopes-da-Silva, J.A., Remize, F., Barba, F.J., Marszalek, K., Delgadillo, I., Saraiva, J.A. (2019): A microbiological, physicochemical, and texture study during storage of yoghurt produced under isostatic pressure. *LWT Food Science and Technology* 110, 152-157. https://doi.org/10.1016/j.lwt.2019.04.066

- Wang, M.L., Morris, B., Tonnis, B., Davis, J., Pederson, G.A. (2012): Assessment of oil content and fatty acid composition variability in two economically important *Hibiscus* species. *Journal of Agricultural and Food Chemistry* 60 (26), 6620-6626. http://doi.org/10.1021/jf301654y
- 42. Watawana, M.I., Jayawardena, N., Waisundara, V.Y. (2018): Value-added Tea (*Camellia sinesis*) as a functional food using the Kombucha 'Tea Fungus'. *Chiang Mai Jornal of Science* 45(1),136-146.
- Wei, J., Ding, W., Zhao Y.G., Vanichpakorn, P. (2011): Acaricidal activity of Aloe vera L. leaf extracts against *Tetranychus cinnabarinus* (Boisduval) (Acarina: Tetranychidae). Journal of Asia Pacific Entomology 14 (3), 353-356.
- **44.** Yerlikaya, O., Kınık, Ö., Akbulut, N. (2011): Milk-derived bioactive peptites and their functional properties. *Journal of Agriculture Faculty of Ege University* **48** (1), 77-84.
- 45. Yao, S., Xie, S., Jiang, L., Li, L. (2017): Effect of dandelion extract, sucrose and starter culture on the viscosity, water-holding capacity and pH of plain yoghurt. *Mljekarstvo* 67 (4), 305-311.
- **46.** Yildiz, O., Eyduran, S.P. (2009): Functional components of berry fruits and their usage in food technologies. *African Journal of Agricultural Research* 4, 422-426.
- Yuksel, Z., Avci, E., Erdem, Y.K. (2010): Characterization of binding interactions between green tea flavanoids and milk proteins. *Food Chemistry* 121, 450-456. https://doi.org/10.1016/j.foodchem.2009.12.064