Investigation of the effect of using carob molasses pulp (CMP) on physicochemical, functional and sensory properties of yogurt

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Abstract
Yogurt, one of the fermented dairy products, is one of the foods consumed daily in many countries. In recent years, many studies have been carried out to increase the nutritional value of yogurt. In this study, a product favored by consumers has been developed by adding carob molasses pulp (CMP), which is rich in antioxidant components and dietary fiber, to yogurt. For this purpose, the effect of adding different amounts of carob molasses pulp (CMP) to yogurt on the physicochemical, chemical and sensory properties of yogurt was investigated. As a result of the yogurt production, it was determined that the pH of the samples decreased (p<0.05) due to the increasing use of CMP, while the total titratable acidity increased (p>0.05). According to the results of color analysis, L* and a * values decreased (p<0.05) and b* values increased (p>0.05) in the samples compared to those with and without CMP. Depending on the use of CMP, significant increases were observed in the antioxidant capacity and total phenolic substance amounts of the samples and it was determined that CMP provided functionality to the final product (3.16 - 5.86 µmol Trolox/100 g and 8.07 - 14.07 mg GAE)/100 g ). According to the results of the sensory analysis, the samples enriched with 0.75% CMP by the panelists in terms of color and appearance, smell, taste, texture and consistency (with a spoon), structure and consistency (in the mouth) and general acceptability received the highest score.

Keywords:
Yogurt, Carob Molasses Pulp, Functional Properties

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INTRODUCTION

In the agriculture-food sector, the dairy industry has an important place in the world (1). Yogurt is the most consumed and preferred fermented fresh dairy products in many countries (2). According to the Turkish Standards 1330 (2006), yogurt is produced by the pasteurization of cow’s milk, sheep’s milk, buffalo milk, goat’s milk or their mixtures, or whether pasteurized milk is homogenized with the addition of milk powder when necessary, *Lactobacillus delbrueckeii* subsp. *bulgaricus* and *Streptococcus thermophilus* are defined as the product obtained after addition of yogurt culture and appropriate processing (3). These cultures produce lactic acid from lactose by fermentation, lowering the pH of the milk from 6.3-6.5 to below 4.6, providing the characteristic taste and odor of yogurt, and providing stability and viscosity to the final product by forming curd firmness (4). A rapid increase in yogurt consumption has been detected in our country and in the world, and this increase is associated with health benefits and functional properties of yogurt such as reducing cholesterol, protecting against cancer, immunological stimulation and improving gastrointestinal functions (5). In addition, yogurt is an important source of nutrients such as protein, fat and minerals for children and adults (6). It has been determined in studies that adding natural food additives with functional effects to the yogurt formulation improves the technological, nutritional and sensory properties of the final product (7, 8, 9).

In addition, carob fruit contains glutamic acid (12.14-12.38 g/100g), alanine (11.15-11.39 g/100g) and aspartic acid (10.76-10.96 g/100g). The total phenolics (13.51 mg GAE/g dry weight), proanthocyanidin (0.36 mg GAE/g dry weight), gallotannins 0.41 CE/g dry weight, total antioxidant activity 145-211 μmol Trolox/g dry weight (12.35). Carob is used in many areas of the food industry due to its rich nutritional composition and functional properties (14). Carob flour, carob gum (locust bean gum), carob molasses and pulp are among these products (15).

One of the important tastes of Turkish culture, molasses is produced by evaporation of the extract obtained by the extraction of carob fruit with water by traditional and industrial methods, up to 70% brix (16, 17). In the production of carob molasses, the part that comes out as waste after extraction is called carob molasses pulp (CMP) (14). CMP contains bioactive components significant for health (18). Since carob molasses pulp has rich in dietary fiber, vitamins, minerals, antioxidants and phenolic components, it is added into food formulations and has an enriching effect on the nutritional and functional properties of the final products (18, 19, 20).

The aim of this study is to produce yogurt rich in nutritional properties. For this purpose, it is aimed to examine the effects of different concentrations of natural carob molasses pulp (0.25%, 0.50% and 0.75%) on the physicochemical, functional and sensory properties of yogurt.

MATERIALS AND METHODS

Raw Materials

Raw CMP was obtained from the company (Ataşıri A.Ş.) that produces carob molasses in Mersin. Firstly, CMP was dried (48°C, 9 hours) and then ground with a laboratory mill. After grinding, the pore diameter was sieved to obtain 100 μm CMP flour.

Yogurt Production

The flow chart of the yogurt production process is given in the figure 1.
YOGURT CHEMICAL ANALYSIS

pH and total titratable acidity analysis

The pH analyses of yogurt samples with CMP and without CMP were measured with a digital pH meter (Ohaus, ST 300) immediately after the yogurts were removed from +4°C, and the pH electrode was washed with pure and immersed in the yogurt samples (21).

Titratable acidity of yogurt samples with CMP and without CMP were calculated as % lactic acid. Samples weighing 10 g were added to 10 mL of distilled water and homogenized. The pH probe was immersed in the sample 20 and titrated with 0.1 N sodium hydroxide until the pH reached 8.1 (21).

Antioxidant activity analysis

Antioxidant potential analysis was detected by the DPPH method. The analysis was carried out using DPPH (1,1-diphenyl-2-picrylhydrazil radical), which can measure the ability to inhibit free radicals, and according to the measurement results of the reaction in methanol against time at 515 nm in the UV-VIS spectrophotometer (22). The antioxidant value of the extracts is expressed as gram Trolox equivalents per 100 g CMP dry weight (gTE 100 gDW⁻¹).

Total phenolic compounds

Total phenolic compounds were determined according to the Folin-Ciocalteau method described by Singleton and Orthofer (23) with some modifications. Firstly, the extraction of the yogurt samples was carried out. Briefly, a sample of 1.0 g was weighed and homogenized with 10 mL of the solvent extraction solution (methanol/distilled water; 80:20 v/v) 80% methanol and centrifuged at 4500 g for 15 minutes, filtered with cellulose filter paper and used for analysis. Prepared sample extracts and Folin-Ciocalteau reagent were used. 0.2 mL of extract was mixed with 1.5 mL of Folin-Ciocalteau reagent (reagent: water mixture 1:10 v/v), respectively, and kept in the dark for 5 minutes. Then, 1.5 mL of saturated sodium carbonate solution and 4 mL of distilled water were added, and the mixture was kept in the dark for about 1 hour at room conditions and its absorbance was measured at 765 nm in a UV-VIS spectrophotometer. The results were evaluated by calculating the phenolic content over the gallic acid equivalent.

![Figure 1. Yogurt production flow chart.](image)

*Commercial yogurt culture label information is indicated that St. thermophilus and Lactobacillus delbrueckii ssp. bulgaricus (L. bulgaricus)
Color Analysis

Color analysis of yogurt samples with CMP and without CMP (PCE, CSM1) were performed with a colorimeter. After the device was calibrated, the samples were placed in a suitable container for solid samples and studied in three parallels and L* (lightness), b* (blueness-yellowness) and a* (greenness-redness) characteristics were determined (24).

Sensory Evaluation

In sensory evaluation of yogurt samples with CMP and without CMP were produced with the use of CMP in different proportions, scoring analysis was performed with the help of a 5-point scale for each predetermined feature, and the result was shown in the spider web diagram (25). The sensory analysis was carried out by a panelist group consisting of 11 academic staff from Toros University. Evaluators rated the yogurts between 1 (very bad) and 5 (very good). Panelists were also given water to clean their mouths before and after tasting each sample.

Statistical Analysis

The findings obtained from the analysis results were evaluated statistically using the SPSS (Version 20, IBM, USA) program. ANOVA was used to compare the means of the obtained data, and the confidence level was set to 95%.

RESULTS

pH and Total Titratable Acidity Values of Yogurt

The pH values of yogurt samples with CMP and without CMP are given in Table 1. As seen in Table 1, the pH varied between 3.77-3.87, and it was obtained that the addition of CMP decreased the pH values of the yogurts compared to the control group (p<0.05). In our research, it was concluded that the addition of CMP to the yogurt formulation activated the metabolic activities of probiotic bacteria and improves the acidity development of the final product (26). In the studies investigating the effect of carob products (flour, extract) on pH of yogurt, it was stated that the pH values were in the range of 3.52-4.97, respectively (21, 27, 28). The pH values measured in our study are similar to the literature. The total acidity in yogurt samples varied between 0.98% (control)-1.03% (CMP75). As the CMP usage rate increased in the samples, the acidity increased and this increase was found to be statistically insignificant (p>0.05). When the literature studies were examined, it was seen that the titration acidity of yogurt samples enriched with carob molasses at different concentrations increased and this situation was associated with rapid protein degradation (27, 29).

Total Phenolic Compounds and Antioxidant Activity of Yogurts

The total phenolic content of the yogurt samples was given in Table 2. It was concluded that CMP added to yogurts significantly increased the total phenolic content of samples (p<0.05). The highest phenolic content values were obtained as 1.03 mg GAE/100 g (CMP75) while the lowest phenolic content value was 0.88 mg GAE/100 g (Control). Functional yogurt was produced due to increased use of CMP in the samples and it was seen that the total phenolic content increased. Carob pod contains a significant amount of polyphenolic compounds (30). In the studies investigating the effect of carob products (flour, extract) on pH of yogurt, it was stated that the pH values increased. Carob pod contains a significant amount of polyphenolic compounds (30).

Table 1. pH and total titratable acidity of yogurts

<table>
<thead>
<tr>
<th>Sample Number</th>
<th>CMP %</th>
<th>pH</th>
<th>Total Acidity (%)</th>
<th>Titrable</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0</td>
<td>4.21±0.01&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.98±0.01&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>CMP25</td>
<td>0.25</td>
<td>4.11±0.01&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.00±0.02&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>CMP50</td>
<td>0.50</td>
<td>4.09±0.01&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.01±0.02&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>CMP75</td>
<td>0.75</td>
<td>4.06±0.58&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.03±0.04&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
</tbody>
</table>

Different superscripts are statistically different in the same column (p<0.05).
nolic compounds (30). The phenolic compounds found in the carob pod are in different subcategories, including phenolic acids (gallic acid, ferulic acid), flavonoids (catechin, epicatechin, myricetin, quercetin) and tannins (digalloyl-glucose, trigalloyl-glucose and tannic acid) (31). Polyphenols can protect cellular components from oxidative damage and reduce the risk of various degenerative diseases (32). For this reason, carob and derived products (powder, syrup, pod) are called functional compounds and are used for enrichment of products (33). Likewise, Kulcan et al. (34) reported that total phenolic compounds of yogurt enriched with additional 12% (w/w) carob extract was 643.46 mg GAE/kg-1246.78 mg GAE/kg, respectively. In another study the effect of carob flour (0-4%) on total phenolic content of yogurts was investigated. The researchers measured the phenolic content of the samples as 27.94 -309.12 µg GAE/g, respectively (21). According to the results, it was determined that carob flour increased the phenolic substance content of yogurt and added functionality to final product.

The antioxidant activity of the yogurt samples was given in Table 2. It was concluded that CMP added to yogurts significantly increased the antioxidant activity of samples (p<0.05). The highest antioxidant activity values were obtained as 3.87 µmol Trolox/100 g (CMP75) while the lowest antioxidant activity value was 3.77 µmol Trolox/100 g (Control). Srour et al. (35) obtained carob-based milk enriched with carob powders. They reported that the antioxidant activity values of the samples varied between 3.5-12.2 mmol Trolox equivalent/kg, respectively. Radia et al. (21) similar to our study, it was reported that the antioxidant capacity values of yogurts that were enriched with carob extract (0-4%) were increased. The high antioxidant activity observed in yogurt samples may have resulted from the reaction between the phenolic compounds provided by carob and organic acids formed as a result of fermentation.

### Color Measurement of Yogurts

Color characteristics of foods directly affect consumer preference and appreciation. The color values (L*, a*, b*) of the yogurt samples enriched with CMP were given in Table 3. The use of CMP in increasing concentration decreased the lightness (L*) and redness (a*) but increased yellowness (b*) of the yogurt samples (p<0.05). As a result, brown yogurts were obtained. The change in color parameters (L*, a*, b*) compared to the control group is explained by the effect of color pigments in CMP (28). Similar to our research, functional yogurt production was carried out using 2% carob powder (36). Researchers have found that carob powder affects the color properties of yogurt, resulting in reduced clarity and intensification of redness and yellowness. Additionally, Scibisz et al. (37) stated that process temperature, acidity, water activity, light exposure and fermentation can affect the pigment content and cause color change in yogurt. In this context, the use of CMP caused color changes in the texture compared to the control group in our research.

### Table 2. The total phenolic compounds and antioxidant activity of yogurts

<table>
<thead>
<tr>
<th>Sample Number</th>
<th>CMP %</th>
<th>Total phenolic compounds (mg GAE/100 g)</th>
<th>Antioxidant activity (µmol Trolox/100 g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0</td>
<td>0.88±0.01c</td>
<td>3.77±0.58d</td>
</tr>
<tr>
<td>CMP25</td>
<td>0.25</td>
<td>1.00±0.02b</td>
<td>3.81±0.08c</td>
</tr>
<tr>
<td>CMP50</td>
<td>0.50</td>
<td>1.01±0.02b</td>
<td>3.83±0.01b</td>
</tr>
<tr>
<td>CMP75</td>
<td>0.75</td>
<td>1.03±0.04a</td>
<td>3.87±0.05a</td>
</tr>
</tbody>
</table>

Different superscripts are statistically different in the same column (p<0.05).
Sensory Properties of Yogurt

Sensory evaluation is one of the best ways to evaluate and accept the final product among consumers (38). The scores of hedonic sensorial qualities of yogurts in terms of color and appearance, texture and consistency (with spoon), texture and consistency (in mouth), taste, and overall acceptability were presented in Table 4. As seen in Table 4, it was concluded that the yogurt samples enriched with CMP were not statistically different from the control sample. The use of increased CMP significantly affected only the color and appearance of yogurts (p<0.05). In our study, it was observed that the use of CMP in yogurt improved the color and appearance properties and was liked by the panelists (p<0.05). This is thought to be due to the unique color characteristic of carob. When the sensory analysis results were evaluated in general, the usage of CMP (0.25-0.75%) in yogurt improved the sensory properties of the product and received high scores from the panelists.

DISCUSSION AND CONCLUSION

In this study, the effect of CMP, which contains phenolic compounds and antioxidant capacity as well as high dietary fiber, on the physicochemical, functional and sensory properties of yogurt was investigated. With the usage of CMP in increasing concentrations in yogurt, slight decrease in the pH value and partial increase in the acidity level were observed, which resulted in an increase in its probiotic properties. In addition, due to the fact that CMP contains bioactive components such as polyphenolic compounds, yogurt has high antioxidant capacity and nutritional fiber content. Thus, yogurt has become more beneficial for human health. The yogurts with CMP added were not different from the control sample in terms of sensory properties except color and appearance. Considering the fact that addition of CMP does not adversely affect the sensory properties of yogurts and taking into account of the high consumer taste, it has been concluded that the usage of CMP from 0.25% to 0.75% is appropriate. As a result, it has been revealed that CMP is an alternative component in the

Table 3. Color parameters of yogurts

<table>
<thead>
<tr>
<th>Sample Number</th>
<th>CMP %</th>
<th>L*</th>
<th>a*</th>
<th>b*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0</td>
<td>82.23±0.51a</td>
<td>6.83±0.01a</td>
<td>9.81±0.03d</td>
</tr>
<tr>
<td>CMP25</td>
<td>0.25</td>
<td>65.71±0.59b</td>
<td>5.16±0.03b</td>
<td>10.21±0.02c</td>
</tr>
<tr>
<td>CMP50</td>
<td>0.50</td>
<td>61.68±0.58c</td>
<td>4.72±0.05c</td>
<td>10.81±0.02b</td>
</tr>
<tr>
<td>CMP75</td>
<td>0.75</td>
<td>60.44±0.51d</td>
<td>3.36±0.04d</td>
<td>11.59±0.01a</td>
</tr>
</tbody>
</table>

Different superscripts are statistically different in the same column (p<0.05).

Table 4. Sensory properties of yogurts

<table>
<thead>
<tr>
<th>Sample Number</th>
<th>CMP %</th>
<th>Color and appearance</th>
<th>Texture and consistency (with spoon)</th>
<th>Texture and consistency (in mouth)</th>
<th>Taste</th>
<th>Odor</th>
<th>Overall acceptability</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0</td>
<td>3.33±0.57b</td>
<td>3.49±0.57a</td>
<td>3.27±0.03a</td>
<td>3.88±0.10a</td>
<td>3.85±0.03a</td>
<td>4.00±0.02a</td>
</tr>
<tr>
<td>CMP25</td>
<td>0.25</td>
<td>4.66±0.58a</td>
<td>4.18±0.16a</td>
<td>3.28±0.12a</td>
<td>4.01±0.01a</td>
<td>3.87±0.12a</td>
<td>3.95±0.06a</td>
</tr>
<tr>
<td>CMP50</td>
<td>0.50</td>
<td>4.66±0.57a</td>
<td>3.50±0.10a</td>
<td>3.26±0.03a</td>
<td>4.00±0.001a</td>
<td>4.00±0.03a</td>
<td>3.95±0.06a</td>
</tr>
<tr>
<td>CMP75</td>
<td>0.75</td>
<td>4.00±0.23a</td>
<td>4.33±0.57a</td>
<td>3.20±0.02a</td>
<td>4.00±0.01a</td>
<td>4.00±0.02a</td>
<td>4.00±0.01a</td>
</tr>
</tbody>
</table>

Different superscripts are statistically different in the same column (p<0.05).
production of functional yogurt and it has improving effect on the physicochemical and sensory properties of yogurt. Additionally, in our study, CMP that was discarded as animal feed or waste was evaluated and it was seen through the conducted analyses that it increased the quality of the final product. Therefore, it may be used as a component of CMP yogurt in the food industry for increasing the health benefits and economic value of products. It may also open the doors to new opportunities for farmers.

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