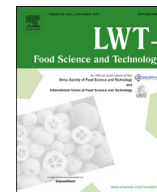




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## Preliminary assessment of a yoghurt-like product manufactured from hazelnut slurry: Study using response surface methodology

Huri Ilyasoğlu<sup>a,\*</sup>, Firat Yılmaz<sup>b</sup>, Nesibe Arslan Burnaz<sup>a</sup>, Cemalettin Baltacı<sup>b</sup>

<sup>a</sup> Department of Nutrition and Dietetics, Gumushane University, Baglarbasi, 29100 Gumushane, Turkey

<sup>b</sup> Department of Food Engineering, Gumushane University, Baglarbasi, 29100 Gumushane, Turkey

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## ABSTRACT

The aim of this study was to evaluate the possibility of using hazelnut slurry in manufacture of yoghurt. A yoghurt-like product was prepared from hazelnut slurry fortified with skimmed milk powder. The effects of the total solids content of the hazelnut slurry (TSCHS, 8–16 g 100 g<sup>-1</sup>) and the content of milk powder (CMP, 6–9 g 100 g<sup>-1</sup>) on the proximate composition, physicochemical and sensorial properties, fatty acid composition, total phenolic content (TPC) and antioxidant activity of the product were evaluated using response surface methodology. Both the TSCHS and the CMP had a significant effect on the total solids content, *b* value, syneresis, palmitic and oleic acid content, and FRAP value. Only the TSCHS showed a significant effect on the protein and fat content, *a* value, water-holding capacity, TPC and ABTS values. Only the CMP showed a significant effect on the carbohydrate and ash content and the acidity. The characteristics of the product generally appeared to be compatible with those of yoghurt. The product was rich in unsaturated fatty acids. Therefore, using hazelnut slurry in manufacture of yoghurt may be proposed to enhance the health benefits of yoghurt.

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### 1. Introduction

Yoghurt, a fermented milk product, is one of the most popular dairy products worldwide because of its nutritional and health benefits. Yoghurt is a rich source of carbohydrate (lactose), protein (casein), fat, vitamins (B vitamins) and minerals (calcium and phosphorus). Yoghurt is an easily digestible product because milk protein, carbohydrate and fat are hydrolysed during fermentation. Yoghurt includes lactic acid bacteria, which have health-promoting properties or therapeutic effects on gastrointestinal functions and diseases, including lactose intolerance, diarrhoea, colon cancer and inflammatory bowel disease. Yoghurt is known to improve bone health and to help control body weight (Adolfsson, Meydani, & Russell, 2004; Mckinley, 2005). Yoghurt is generally manufactured from dairy milk, especially cow's milk. Many attempts have been made to produce yoghurt from plant milk including soy milk (Granata & Morr, 1996; Rinaldoni, Campderros, & Padilla, 2012), mango-soy milk (Kumar & Mishra, 2004), corn milk

(Supavititpatana, Wirjantoro, Apichartsrangkoon, & Raviyan, 2008) and peanut milk (Isanga & Zhang, 2009).

Hazelnuts are important in human nutrition and health because of their composition of protein, carbohydrates, lipids, vitamins, minerals, dietary fibres, tocopherols, phytosterols, squalene, and phenolic compounds (Alasalvar, Shahidi, Liyanapathirina, & Ohshima, 2003). Epidemiological studies have shown that nut consumption is associated with a lower risk of coronary heart disease. Nut consumption has also been shown to help prevent sudden cardiac death, hypertension, gallstone disease, high blood cholesterol and high blood pressure (Ros, 2010). Hazelnut slurry is produced by soaking hazelnuts (roasted or unroasted) in water, grinding the nuts in water and then filtering the slurry. Hazelnut slurry may provide the same potential health benefits as hazelnuts. Hazelnut slurry is rich in monounsaturated fatty acids (mainly oleic acid) and phytosterols (mainly  $\beta$ -sitosterol) and contains antioxidant compounds.

Hazelnut slurry may be used in the manufacture of yoghurt to enhance the health benefits of yoghurt. The aim of this study was to evaluate the possibility of using hazelnut slurry in manufacture of yoghurt. The effect of the ingredients on the characteristics of the product was analysed using response surface methodology.

\* Corresponding author. Tel.: +90 456 233 74 25 1865; fax: +90 456 233 76 04.

E-mail addresses: [huriilyasoglu@yahoo.com](mailto:huriilyasoglu@yahoo.com), [hilyasoglu@gumushane.edu.tr](mailto:hilyasoglu@gumushane.edu.tr) (H. Ilyasoğlu).

## 2. Material and methods

### 2.1. Chemicals

All chemicals and solvents (analytical grade or HPLC grade) were obtained from Merck (Darmstadt, Germany). FAME mix, ABTS, TPTZ and gallic acid were purchased from Sigma-Aldrich (St. Louis, MO, USA).

### 2.2. Materials

Hazelnuts (Tombul cultivar) were obtained from an orchard in Giresun province (Turkey). Skimmed milk powder (Pinar Co., Izmir, Turkey) was purchased from a local market. Skimmed milk powder had protein (36 g 100 g<sup>-1</sup>) and carbohydrate (52 g 100 g<sup>-1</sup>) as the main constituents. The starter culture (Chr. Hansen FD DVS YC-X16, Chr. Hansen A/S, Horsholm, Denmark) was obtained from a local distributor.

### 2.3. Hazelnut slurry preparation

Shelled hazelnuts were roasted at 140 °C for 15 min in an oven. The roasted hazelnuts were soaked in water for 12 h. After filtration and washing, the hazelnuts were ground with water in a blender (Waring laboratory blender, Conair Corporation, Stamford, CT, USA) for 2 min. The slurry was filtered through a double-layered cheesecloth.

One batch of the hazelnut slurry was prepared and the total solids content of the batch was determined. When hazelnuts were ground with water (1:3) in a blender, the obtained hazelnut slurry had approximately 24 g 100 g<sup>-1</sup> of total solids content. It had 11.6 g of fat, 7.4 g of carbohydrate and 4.6 g of protein as the main constituents. The batch was diluted with water to obtain the targeted levels of total solids.

### 2.4. Yoghurt manufacture

The skimmed milk powder was dissolved in the hazelnut slurry at 43 °C, stirring for 40 min. The milk was homogenised with a homogeniser (Daihan WiseTisHG-15A, Daihan Scientific Co., Seoul, South Korea) and pasteurised at 90 °C for 20 min. After cooling to 43 °C, the starter culture (3 mL 100 g<sup>-1</sup>) was added to the pasteurised milk. The milk inoculated with the starter culture was incubated at 43 °C for 4–4.5 h until a pH of about 4.6–4.7 was attained. The yoghurt was stored at 4 °C overnight prior to analysis.

### 2.5. Proximate composition

The moisture, protein, fat and ash content of the samples were determined in accordance with the AOAC methods. The oven method was used for the moisture content, the Kjeldahl method for the protein content (factor: 6.38), the Gerber method for the fat content, and the dry burning method for the ash content. Total carbohydrates were calculated by subtracting the total percentages of moisture, protein, fat and ash from 100.

### 2.6. Physicochemical properties

The pH of the sample was measured with a pH meter (Hanna HI 2210, Smithfield, RI, USA). The acidity of the sample was determined by the alkali titration method. Colour properties (*L*, *a*, and *b* values) were measured with a chromometer (Konica Minolta CR-400, Japan). The syneresis was measured by using 10 g of yoghurt spread on a filter paper (Whatman No. 1) in a beaker. The beaker was held at 4 °C for 5 h, and the liquid collected was weighed. The

water-holding capacity was determined with the centrifuge method. The yoghurt sample (10 g) was stored at 4 °C for 24 h, and then the tubes were centrifuged at 5000 × g for 20 min at 4 °C. The whey separated from the samples was weighed.

### 2.7. Fatty acid composition

The fatty acid composition was determined according to the analytical methods previously described (Ilyasoglu, 2013). The determination of the fatty acid composition was carried out by gas chromatography with flame ionisation detection (GC-FID). Fatty acid methyl ester (FAME) was injected into a Shimadzu GC-2010 Plus gas chromatograph (Shimadzu Corporation, Japan) equipped with a flame ionisation detector, a split/splitless injector and a long capillary column (0.25 mm × 0.20 μm × 60 m, Teknokroma TR-CN100, Teknokroma Anlitica, Barcelona, Spain). The oven temperature program was as follows: the initial temperature of the column was 90 °C, held for 5 min, followed by a 10 °C/min ramp to 240 °C, and held for 20 min. The carrier gas was helium at a flow rate of 1 mL/min, the split ratio was 50:1, and the injection quantity was 1 μL. The identification of FAMES was performed by using a standard FAME reference mixture. The peak areas were computed by the integration software, and fatty acids were given in percentages relative to the total fatty acid content.

### 2.8. Total phenolic content

The samples (2 g) were extracted with 5 mL of methanol (70%) for one hour in an ultrasonic bath and centrifuged for 10 min. After filtration, the residue was re-extracted with 5 mL of methanol (70%). The combined methanol extracts were stored at -18 °C until analysis. The total phenolic content (TPC) was estimated using the Folin-Ciocalteu method. A total of 0.1 mL of the extract solution was mixed with 0.50 mL of diluted Folin-Ciocalteu reagent, 0.4 mL of sodium carbonate (1 M) and 4 mL of distilled water. The absorbance of the mixture was measured at 760 nm after 1 h. The calibration curve was prepared with standard gallic acid ranging from 0 to 200 mg/mL. The TPC was expressed as mg of gallic acid equivalents (GAEs) per kg of the sample.

### 2.9. ABTS radical scavenging activity

For the ABTS assay, the ABTS stock solution was prepared by reacting 7 mmol/L of ABTS with 2.45 mmol/L of potassium persulphate solution. The solution was then left in the dark at room temperature for 16 h. The stock solution was diluted with ethanol to reach an absorbance of 0.70 (±0.02) AU at 734 nm. A total of 50 μL of the extract was mixed with 1500 μL of ABTS<sup>+</sup> solution, and the absorbance was measured at 734 nm after 6 min. The results were expressed as micromoles of trolox per kg of the sample.

### 2.10. Ferric reducing antioxidant power assay

For the ferric reducing antioxidant power (FRAP) assay, fresh FRAP reagent was prepared by mixing the following solutions (10:1:1): acetate buffer solution (pH = 3.6), TPTZ solution in 40 mmol/L HCl (10 mmol/L) and FeCl<sub>3</sub> (20 mmol/L) solution. A total of 50 μL of the extract was mixed with 1500 μL of FRAP reagent, and the absorbance was measured at 595 nm after 20 min. The results were expressed as micromoles of trolox per kg of the sample.

### 2.11. Microbiological analyses

A check of starter culture in the samples was performed immediately after the completion of fermentation and during four

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