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(Article begins on next page)

INFLUENCE OF THE ADDITION OF DIFFERENT HAZELNUT SKINS ON THE PHYSICOCHEMICAL, ANTIOXIDANT, POLYPHENOL AND SENSORY PROPERTIES OF YOGURT

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Abstract

Skins obtained from three different varieties (Georgia, San Giovanni and Tonda Gentile Trilobata) of roasted hazelnuts (*Corylus avellana* L.) were used at two different percentages (3% and 6%) in yogurt

production to increase the dietary fibre and polyphenol content. The effects on the physico-chemical characteristics, antioxidant capacity, phenolic compounds, and sugar and organic acid content during 3 weeks of storage at 4 °C were evaluated, and a preference test was performed with consumers at the end of storage.
The amount of skin and the variety used significantly influenced all of the physico-chemical parameters and were associated with consumer preference. Concerning the dietary fibre content, total polyphenol content and antioxidant capacity, all of which affect the functional ability of food products, the highest values obtained were for all of the products contained a hazelnut skin content of 6%. Among the cultivars, the highest values obtained were for yogurt with the Georgia hazelnut skin. Although 6% hazelnut skin yogurts displayed the highest functional ability, a decreased consumer preference was observed; yogurt with 3% San Giovanni and Tonda Gentile Trilobata hazelnut skins had the maximum consumer rating.

1. Introduction

The production of hazelnuts in 2012 was 914.447 $*10^9$ kg. Turkey was the world's largest producer and contributed 72% of the total production, followed by Italy (9.3%), the United States (3.3%) and Georgia (2.7%) (FAOSTAT, 2012). Two different by-products are obtained during the transformation of hazelnuts through the post-harvesting processes - shells and hazelnut skin - among these, only the shell has a direct commercial value as a heating source. Hazelnut skin, representing approximately 2.5% of the total kernel weight (Alasalvar et al., 2009), is a rich source of dietary fibre as well as phenolic compounds with antioxidant properties (Del Rio, Calani, Dall'Asta, & Brighenti, 2011). The definition of dietary fibre and its beneficial effects on human health has been considerably debated and related to physiological considerations (EFSA, 2010). Dietary fibre is categorized into two groups according to water solubility: water-soluble dietary fibre (SDF) and water-insoluble dietary fibre (IDF). SDF forms a viscous solution that results in increased viscosity in the intestine, leading to slowed intestinal transit, delayed gastric emptying and slowed glucose and sterol absorption, whereas

IDF has a high water-holding capacity that contributes to increased faecal bulk. Currently, an average daily fibre intake of 25 g for adults and 10 g (1-3 years old) to 21 g (17 years old) for children is recommended.

Antioxidants are notably important compounds in food science due to their ability to prevent lipid oxidation in foods and to decrease the negative effects of reactive oxygen species on physiological functions in humans. Polyphenols, which are widely distributed in plants, are among the most studied natural antioxidants due to consumer preference for natural products. Currently, a daily polyphenol intake of 1 gram is reported (Scalbert, Manach, Morand, Rémésy & Jiménez, 2005). Recently, hazelnut skin itself or its phenolic extracts have been added to vanilla ice cream, bread or coffee to investigate the effects on the final products in terms of fat replacement, as a source of dietary fibre and as a potential source of antioxidants, respectively.. The application of hazelnut skin to ice cream demonstrated that it could improve product overrunning, but it resulted in greater susceptibility to melting and was not preferred by consumers (Dervisoglu, 2006). The use of hazelnut skin in bread revealed that a concentration of 5% did not considerably affect the rheological properties of the dough or the final product and produced acceptable results from the sensory panel (Anil, 2007). Contini, Baccelloni, Frangipane, Merendino, and Massantini (2012) emphasized that phenolic extracts from hazelnut skins increased the antiradical activity of coffee due to an increase in the total polyphenol content.

Therefore, the aim of this work was to evaluate the possibility of using hazelnut skin as a source of dietary fibre and antioxidants in yogurt. The use of hazelnut skin in yogurt could have a dual benefit by employing a food industrial by-product for human nutrition, thereby reducing industrial waste. In addition, it could augment the consumption of fibre and antioxidant compounds in all sectors of the population owing to the popularity of yogurt around the world (61.248 $*10^9$ kg yogurt production - FAOSTAT, 2012).

2. Materials and Methods

2.1. Hazelnut skin (HS) samples

The skins of three different hazelnut (*Corylus avellana* L.) varieties ("Tonda Gentile Trilobata - TGT", "San Giovanni" cultivars from Italy, and "Georgia" from Georgia) were obtained from the Nocciole Marchisio S.p.A. (Cortemilia, CN, Italy). The roasting process was conducted under three different conditions (temperature: 155, 150 and 155 °C; time 37, 35, 39 min, respectively). Conventional procedures were applied by the processor in an industrial continuous-working oven, where the skins were separated from the roasted kernels by vigorously rubbing them against themselves, followed by skin removal via vacuum.

2.2.Chemicals

All reagents and solvents were purchased from Sigma-Aldrich (Milano, Italy). All chemicals were reagent-grade, and ultrapure water was produced with a Milli-Q System (Millipore, Milan, Italy).

2.3. HS preparation

HS were collected just after industrial processing and transported to the laboratory in vacuum bags. HS were milled and sieved to obtain a particle fraction of 0.5 mm using an ultra-centrifugal mill Retsch ZM 200 (Retsch Gmbh, Haan, Germany). The resulting products were stored at 4 °C.

2.4.Chemical composition of HS and fortified yogurt

The moisture content was determined using a Radwag MAC 210/NH thermo-balance (Radwag, Radom, Poland) at 105 °C. The total protein content (conversion factor 6.25) was obtained according to the Kjeldahl method using a UDK 130A system (Velp Scientifica, Usmate, Italy). The lipid fraction was extracted using a Soxhlet Velp Extraction System SER 148 (Velp Scientifica, Usmate, Italy) for 6 h using *n*-hexane as solvent. The ash content was determined in a muffle furnace according to the AOAC (1990) method. The carbohydrate value was estimated by the difference. Dietary fibre (TDT, SDF and IDF) was measured using the Megazyme Total Dietary analysis kit according to the enzymatic gravimetric method proposed by Lee, Prosky, and Devries, (1992). Compositional analyses of fortified yogurt were run 24 hours after yogurt production. All analyses were performed in triplicate.

2.5.Yogurt preparation

A single lot of stirred yogurt was prepared from UHT whole milk (fat 3.6%; protein 3.1% and carbohydrates 4.8%) purchased at the local market. Milk was placed into a vat and allowed to cool at 42 °C and was subsequently inoculated with the starter culture YO-MIX 401 (Santamaria, Burago di Molgora, Italy), which is a combination of *Streptococcus thermophilus* and *Lactobacillus delbrückii* subsp. *Bulgaricus*. Incubation was carried out at 42 °C until the pH was 4.8 (approximately 6.5 h). After the desired pH was reached, the fermentation was interrupted by cooling the vat to 20 °C. The coagulum was then broken with a stainless steel skimmer. The HS content of the yogurt was directly adjusted (0, 3 and 6 g were added to obtain 100 g of yogurt designated as the control 0%, 3% and 6%, respectively) in single pots. Yogurt was kept at 4 °C and analysed on days 1, 7, 14 and 21 of storage.

2.6. Analysis of the physico-chemical characteristics of yogurt

The pH of the samples was measured with a Crison microph 2002 pH-meter (Crison Strumenti SpA, Carpi, Italy). The titratable acidity was determined by the potentiometric method according to the IDF standard (IDF, 1991) and expressed as the lactic acid %. Yogurt syneresis was determined by the centrifugation method of Celik, Bakırcı, and Şat (2006), with several modifications. Twenty grams of yogurt were centrifuged at 16800 × *g* for 20 min at 4 °C using a Megafuge 11 R centrifuge (Thermo Fischer Scientific, Waltham, MA, USA). Syneresis was expressed as the volume of separated whey per 100 mL of yogurt. All of the analyses were performed in triplicate.

2.7.Microbiological analysis

Microbiological analyses of yogurt were performed to determine the influence of the HS addition on the starter. Streptococci were counted on M-17 agar (Oxoid, Basingstoke, Hampshire, England) and were incubated aerobically at 37 °C for 24 h. Lactobacilli were counted on MRS agar (Lab M Limited, Heywood, Lancashire, United Kingdom) under anaerobic incubation at 37 °C for 48 h. The samples were analysed in duplicate.

2.8.Antioxidant capacity of yogurt

2.8.1. *Bioactive compounds extraction*

Yogurt extracts were prepared according to McCue and Shetty (2005), with slight modifications. Briefly, each yogurt sample (10 g) was diluted with distilled water (2.5 ml) and centrifuged (16800 × *g*, 40 min, 4 °C). The supernatant was harvested and filtered through a 0.45-μm polypropylene membrane filter (VWR, Milan, Italy). Extraction was conducted in triplicate, and extracts were stored at 4 °C in amber glass vials until further analyses.

2.8.2. Total phenolic content assay

The total phenolic content (TPC) was determined using the Folin-Ciocalteu assay as reported by Apostolidis, Kwon, and Shetty (2007) after the reaction samples were centrifuged (16800 × *g*, 10 min, 20 °C), and the absorbance of the supernatant was measured at 725 nm with a UV-Visible spectrophotometer (UV-1700 PharmaSpec, Shimadzu, Milan, Italy). The results were expressed as μg gallic acid equivalents (GAE) per gram of sample (calibration curve linearity range: r = 0.997).

2.8.3. DPPH radical scavenging capacity of yogurt

The free radical scavenging activity (RSA) of the extracts was determined according to the procedure reported by von Gadow, Joubert, and Hansmann (1997) using the stable 2,2-diphenyl-1-picrylhydrazyl radical ($DPPH^{\bullet}$). Briefly, 75 μL of sample extract was added to 3 mL of a 6.1×10^{-5}

M DPPH$^{\bullet}$ methanol solution and incubated for 1 h at room temperature in the dark. After this time and after a centrifugation step (16800 × *g*, 10 min, 20 °C), the decrease in absorbance at 515 nm was recorded against methanol as a control; a methanol solution of DPPH$^{\bullet}$ was used as a blank. The inhibition percentage (IP) of the DPPH$^{\bullet}$ by the antioxidant extracts was calculated according the formula

$$IP = [(A_{0min} - A_{60\ min})/A_{0min}] \times 100$$

where A_{0min} is the absorbance of the blank at t = 0 min and $A_{60\ min}$ is the absorbance of the samples at 60 min. The results were expressed as µM Trolox equivalents (TE) per gram of sample by means of a dose-response curve for Trolox (0-350 µM).

2.9.HPLC-DAD Phenolic compound analysis

HPLC-DAD analysis was performed by using a Thermo-Finnigan Spectra-System HPLC system (Thermo-Finnigan, Waltham, USA) equipped with a P2000 binary gradient pump system, a SCM 1000 degasser, an AS 100 automatic injector, an UV6000LP DAD and ChromQuest software for data processing. Separation was achieved on a C_{18} RP Lichrosphere 250 × 4.6 mm, 5-µm (Merck, Milan, Italy) column equipped with a C_{18} RP Lichrosphere 5-µm guard column (Merck, Milan, Italy). The mobile phase was composed of trifluoroacetic acid/ultrapure water (0.1:99.9, v/v) (A) and methanol (B). The flow rate was 1 mL/min, and the injection volume was 20 µL. The elution program was as follows: 95% A as the initial condition, maintained for 2 minutes; 80% A for 8 min; 25% A for 57 min; 0% A for 13 min; and 95% A for 5 min. DAD spectra were recorded in full scan mode over a wavelength range of 200 to 400 nm. Identification was achieved by comparing the retention times and spectra with authentic standards (Fig. 1). Each compound was quantified as mg/Kg sample by means of calibration with external standards: gallic acid, protocatechuic acid, procyanidin B1, gallocatechin gallate, 3-coumaric acid and rutin purchased from Sigma-Aldrich (Milan, Italy) and 2-coumaric acid purchased from Extrasynthese (Genay Cedex, France).

2.10. HPLC-UV-RI Organic acids and sugars analysis

The content of organic acids and sugars was determined according to the method of Adhikari, Grün, Mustapha, and Fernando (2002). The HPLC system (Thermo Quest, San Jose, CA) was equipped with a P4000 isocratic pump, a multiple autosampler AS3000 fitted with a 20-µL loop, a UV detector (UV100) set at 210 nm, and a refractive index detector (Spectra System RI-150, Thermo Electro Corporation). The detectors were connected in series. Data were collected using ChromQuest ver. 3.0 (Thermo Finningan). The mobile phase was 0.01 N H_2SO_4, and the analyses were performed isocratically at 0.8 mL/min and 65 °C with a 300 × 7.8 mm i.d. cation exchange column (Aminex HPX-87H) equipped with a cation H^+ microguard cartridge (Bio-Rad Laboratories, Hercules, CA). Identification was achieved by comparison with the retention times of authentic standards: lactose, glucose, galactose, pyruvic acid, lactic acid, malic acid and citric acid purchased from Sigma-Aldrich (Milan, Italy).

2.11. Preference test

To assess the sensory acceptability of the yogurts, twenty consumers (40% male and 60% female, aged between 24 and 65 years) were recruited at the Dipartimento di Scienze Agrarie, Forestali e Alimentari of Turin University. Written informed consent was obtained from each subject after the experiments were described.

The test was performed inside an air conditioned room with white light at approximately 21 °C. Yogurt samples (10 g) were served blinded in a transparent plastic cup coded with a random three-digit number. Samples were served in a completely randomized order. Consumers were asked to rate their preference for odour, taste, flavour, texture and acceptability. Preference was expressed on a 5-point hedonic scale ranging from “dislike extremely” (1) to “like extremely” (5) (Peryam & Pilgrim, 1957). Paper score-sheets were used for data collection.

2.12. Data analysis

A one-way analysis of variance (ANOVA) with Duncan's test for mean comparison was used to highlight significant differences among the yogurt samples. All calculations were performed with the STATISTICA software for Windows (Release 7.0; StatSoft Inc., Tulsa, OK, USA).

3. Results

3.1.Chemical composition of HS

Table 1 shows the chemical composition of HS. According to the results, total dietary fibre was the major component, amounting to a mean of 55%. A mean of 86% of the fibre was composed of insoluble fibre, with significant differences among the varieties. The lipid content ranged from 109.96 ± 1.68 g/Kg for Georgia samples to 187.55 g/kg for San Giovanni samples. The values were similar to those reported by Anil (2007) as well as Turhan, Sagir and Ustun (2005) for other varieties.

The TPC values assessed in hazelnut skin extracts significantly characterized the varieties. The highest values were measured in the Georgia skin extracts, and the lowest values were found in the San Giovanni skin extracts; nevertheless, there were no significant difference for TGT.

The results of the RSA assays revealed a different trend - the RSA had the highest values reported for the Georgia sample, followed by San Giovanni and TGT.

The use of different extraction methods and/or different data expression methods prevented the comparison of our TPC and RSA results with those published by other authors.

3.2.Chemical composition of yogurt

Table 2 shows the chemical composition of the yogurts. The overall composition of the yogurts was significantly different ($p<0.001$). In particular, yogurt with HS was associated with a mean decreased humidity of 2.9% and 6.0% for the 3% and 6% HS treatments, respectively, but the differences observed among the different varieties were not statistically significant. These results are in accordance with those obtained by García-Pérez, Lario, Fernández-López, Sayas, Pérez-Alvarez and Sendra, (2005) who added citrus fibre to yogurt.

The addition of hazelnut skin was also associated with a decrease in protein, lipids, carbohydrates and ash.

As expected, the addition of HS was associated with the dietary fibre level in the final product. Furthermore, the dietary fibre content increased with the mean values of 94.65 ± 28.19 g/Kg and 165.19 ± 4.91 g/Kg in yogurt with 3% and 6% HS, respectively. Among the varieties, the highest concentration was observed in yogurt fortified with Georgia, but no differences were observed between San Giovanni and TGT cultivar HS. Similar data for total dietary fibre showing an increase in yogurt due to added fibre were obtained by do Espírito Santo et al. (2012) and Tseng and Zhao (2013). The results showed an increase in total dietary fibre for all of the matrices used, and as expected, the fibre content in the final product increased with an increasing percentage of the ingredients studied.

For the soluble and insoluble dietary fibre content, the highest concentrations were observed for both yogurt samples with different percentages of Georgia HS.

3.3.Physico-chemical characteristics of yogurt

The pH, titratable acidity and syneresis of yogurts are reported in Table 3. The pH of all products dropped slightly ($p < 0.001$) during storage independent of the HS addition. Among the products, the 6% Georgia fortified yogurt showed the lowest pH reduction during storage (0.19 unit), while the 6% TGT fortified yogurt had the highest pH reduction (0.28 unit). The mean reduction was 0.24 units and was lower than that reported in other studies in which different types of by-products were added to yogurt (García-Pérez et al., 2005; Tseng & Zhao, 2013), but was slightly higher than that found by others when different pure dietary fibres were added (Dello Staffolo, Bertola, Martino, & Bevilacqua, 2004). Moreover, a significant difference ($p<0.001$) between the types and percentages of HS used was present between the first and the second week of storage, but at the end (3 weeks), only the yogurt with 3% TGT HS was different from the others.

For syneresis, the addition of HS was associated with increased whey separation compared to the control at all storage times ($p<0.001$) due to the rearrangement of the gel matrix being associated with the high content of insoluble dietary fibre in the HS, as previously observed by García-Pérez et al. (2005) and Tseng and Zhao (2013). Among the two percentages of HS, regardless of the varietal used, a difference with a mean value of 9% was observed. Only the Georgia 6% and the TGT 3% fortified yogurts showed significantly different values during storage.

For titratable acidity, the incorporation of HS in the yogurts was associated with statistically significant differences between the products for all storage periods. The 6% TGT fortified yogurt showed the highest increase in acidity during storage (0.81 unit), and the 3% TGT fortified yogurt had the lowest (0.06 unit).

3.4.Microbiological analysis

As shown in Fig. 2, the addition of HS to yogurt did not affect the survival of the starter strains; after 21 days of storage, both strains had a concentration higher than that required by the Codex Alimentarius (10^7 CFU/g). In particular, in the fortified yogurts, *S. thermophilus* reached a mean concentration of 8.67 $\log_{10}$ CFU/mL, which was higher than the control (8.38 $\log_{10}$ CFU/mL). *L. bulgaricus* was present at a mean concentration of 7.73 $\log_{10}$ CFU/mL in fortified yogurt compared to 7.64 $\log_{10}$ CFU/mL in the control.

The viability of *S. thermophilus* decreased during refrigerated storage (Fig. 2 A & B), but by less than 1 CFU/mL. TGT HS was associated with the highest reduction, while the lowest reduction was observed for Georgia 3% and San Giovanni 6%.

The viability of *L. bulgaricus* decreased during refrigerated storage (Fig. 2 C & D), but was less than 1 CFU/mL and less than that observed for the *S. thermophilus*, except for TGT 3% and 6%.

As observed for *S. thermophilus*, TGT HS was associated with the highest reduction in *L. bulgaricus*; the lowest was observed for Georgia 3% and San Giovanni 6%.

3.5. Total phenolic content and antioxidant capacity of yogurt

Table 4 shows the total phenolic content and the free radical scavenging activity of the yogurts. During the storage period, the TPC observed for the control yogurt dropped significantly ($p<0.001$) due to bacterial metabolic activity associated with a reduction/modification of the non-phenolic compound that reacted with the Folin-Ciocalteu reagent (Everette, Bryant, Green, Abbey, Wangila, & Walker, 2010).

Fortified yogurts showed statistically significant differences at each storage time ($p<0.001$), and among the samples, a statistically significant increase was observed during storage. This increase is in accordance with the results obtained by Zainoldin and Baba (2009) for yogurt fortified with dragon fruit, but contrasts with results obtained by other researchers for yogurt fortified with grape pomace (Tseng & Zhao, 2013), different grape berries and callus extract (Karaaslan, Ozden, Vardin, & Turkoglu, 2011) and *Berberis boliviana* anthocyanins (Wallace & Giusti, 2008). Addition of 3% HS increased the total phenolic compound concentrations by 36.5, 29.4, and 27.4% for TGT, Georgia and San Giovanni, respectively. Addition of 6% HS increased the concentration by 30.9, 26.7 and 26.3% for TGT, San Giovanni and Georgia, respectively.

During storage, the RSA of control samples significantly increased ($p<0.005$), possibly because bacterial metabolic activity caused a breakdown of macromolecules that could react with the $DPPH^{\bullet}$ reagent.

Fortified yogurts showed storage trends similar to those observed for TPC. In storage, the addition of 3% HS showed an increased RSA of 41.6, 52.4, and 69.4% for San Giovanni, Georgia, and TGT, respectively, and the addition of 6% HS showed an increased RSA of 30.6, 39.5 and 73.6% for Georgia, San Giovanni and TGT, respectively.

3.6. Phenolic compounds profile

The most abundant phenolic compound was procyanidin B1, followed by protocatechuic acid, gallic acid, gallocatechin gallate, rutin and 3-coumaric acid (Table 5). 2-coumaric was detected only in the Georgia HS samples, but was not quantified. None of the phenolic compounds found in the fortified yogurts were detected in the control samples.

Yogurts with 6% HS showed a higher concentration of phenolic compounds (except for coumaric acid and gallocatechin gallate) than those with 3% HS. The compounds detected were unchanged during storage in almost all samples. An increase in gallic acid (in the San Giovanni and TGT cultivars at both percentages), protocatechuic acid (in the TGT cultivar at 6% HS) and rutin (in San Giovanni cultivar at 3% HS and TGT cultivar at 6% HS) during storage could be attributed to an increase in compound solubilization into the yogurt, probably due to the decrease of pH during storage (Stalikas, 2007), followed by major extraction in water. Statistically significant variations in procyanidin B1 and protocathechuic acid were found among the HS varieties at each sampling time. The lowest concentrations were detected in San Giovanni HS, whereas the highest were observed in Georgia HS. Statistically significant differences for gallic acid were found among the HS varieties at each storage time. The lowest concentration was detected in Georgia HS, while the highest was observed in TGT HS.

The highest rutin concentrations were detected at days 7 and 21 in yogurts with 6% San Giovanni HS, while the lowest were found in yogurts made with 3% San Giovanni and TGT HS (< LOQ).

3.7.Organic acid and sugar profiles

Table 6 shows the sugar and organic acids concentration of the yogurts. No statistically significant differences in the lactose concentration were observed among the samples at any sampling time. The 3% HS was associated with higher lactose degradation, as indicated by a higher bacterial count at each storage time (Fig. 2). Statistically significant differences for the control, Georgia 3% and 6%

and San Giovanni 3% samples were observed, in which lactose degradation was 6.7, 9.0, 7.2 and 6.9%, respectively.

Statistically significant differences for glucose and galactose were observed for both the varieties at each storage time and for each sample during storage, except in the San Giovanni 6% sample. In particular, the control samples evidenced an increase in the galactose concentration of 11.4% during the storage period, while in the other samples, the galactose concentration decreased with a mean percentage of 22.2% and 20.0% for 3% and 6% HS, respectively. The highest degradation was observed in TGT yogurt samples and the lowest in the San Giovanni samples.

An increase in the glucose concentration was observed in the control and the 3% and 6% San Giovanni HS samples during the storage period, amounting to 159.5, 6.4 and 23.3%, respectively. In the other samples, a decrease occurred that amounted to a mean percentage of 43.5 and 120.0% for the 3% and 6% HS samples, respectively. The highest degradation was observed in the Georgia samples and the lowest in the TGT samples.

For citric acids, no significant differences were observed, indicating that starter bacteria do not utilize citrate, possibly because they are a Cit^- strain as previously mentioned by Adhikari, Grün, Mustapha, and Fernando, (2002).

During the storage time, the concentration of pyruvic acid increased. However, this increase was not constant during storage, possibly because it is an intermediary product of bacterial metabolism and its concentration normally fluctuates during storage as a function of bacterial activity. Lactic acid showed a statistically significant increase during storage. Regardless of variety, the mean increase observed was 10.0% and 14.4% for 3% and 6% HS, respectively. Among the varieties, the highest increase was observed in 3% San Giovanni and 6% TGT.

Malic acid was not detected in the control samples because it is an acid derived from HS. Statistical differences were observed between the varieties and the HS levels. As expected, an increased concentration of HS in yogurt was associated with a higher concentration of malic acid. Among the varieties, the highest concentration was detected in the San Giovanni samples and the lowest in the Georgia samples.

3.8.Sensory analysis

Fig. 3 shows the consumer acceptance of yogurts. The fortification of yogurt with the HS was associated with a statistically significant effect ($p<0.001$) on all of the parameters analysed except for odour. The control sample was acceptable. For all of the parameters analysed, the control scored the central value of the scale (3 = neither like nor dislike). Consumers preferred 3% HS to 6% HS. This preference can possibly be explained because HS was associated with increased liquidity of the samples (see syneresis value Table 3).

For the 3% HS samples, the San Giovanni and TGT cultivar scores always achieved the central scale value for the 6% HS samples. The San Giovanni cultivar had the highest score for all of the parameters, but only the odour achieved the central scale value.

In general, the observed low acceptance of the fortified yogurts was not surprising because similar results have been previously observed in other studies in which different types of fibre were used. Tseng and Zhao (2013) observed that the use of fibre was associated with a lower value for flavour, texture and consistency. Hashim, Khalil, and Afifi (2009) reported that the addition of fibre was associated with lower ratings for firmness, smoothness and flavour. Sendra, Fayos, Lario, Fernández-Lopez, Saras-Barberá, and Pérez-Alvarez, (2008) observed that the addition of fibre was associated with reduced creaminess and decreased overall acceptability.

4. Conclusions

This study demonstrated that HS can be utilized as an alternative source of antioxidants and dietary fibre to fortify yogurt. The addition of HS and the percentage added contributes to the dietary fibre content and antioxidant capacity of the final product, as well as to all of the other physico-chemical parameters considered. During storage, the antioxidant capacity of fortified products was increased with respect to the control, and no modification of the phenolic compounds was observed. Thus, it is possible to conclude that the functional ability of these products is stable or increased during storage. The yogurt with the 3% San Giovanni and TGT HS achieved the highest score from the consumers. By consuming 100 g of products fortified with 3% of these two varieties, consumers obtain the 37% dietary fibre intake recommended by the European Union and the respective 0.4 and 0.6 %, polyphenol intake reported by the scientific literature.

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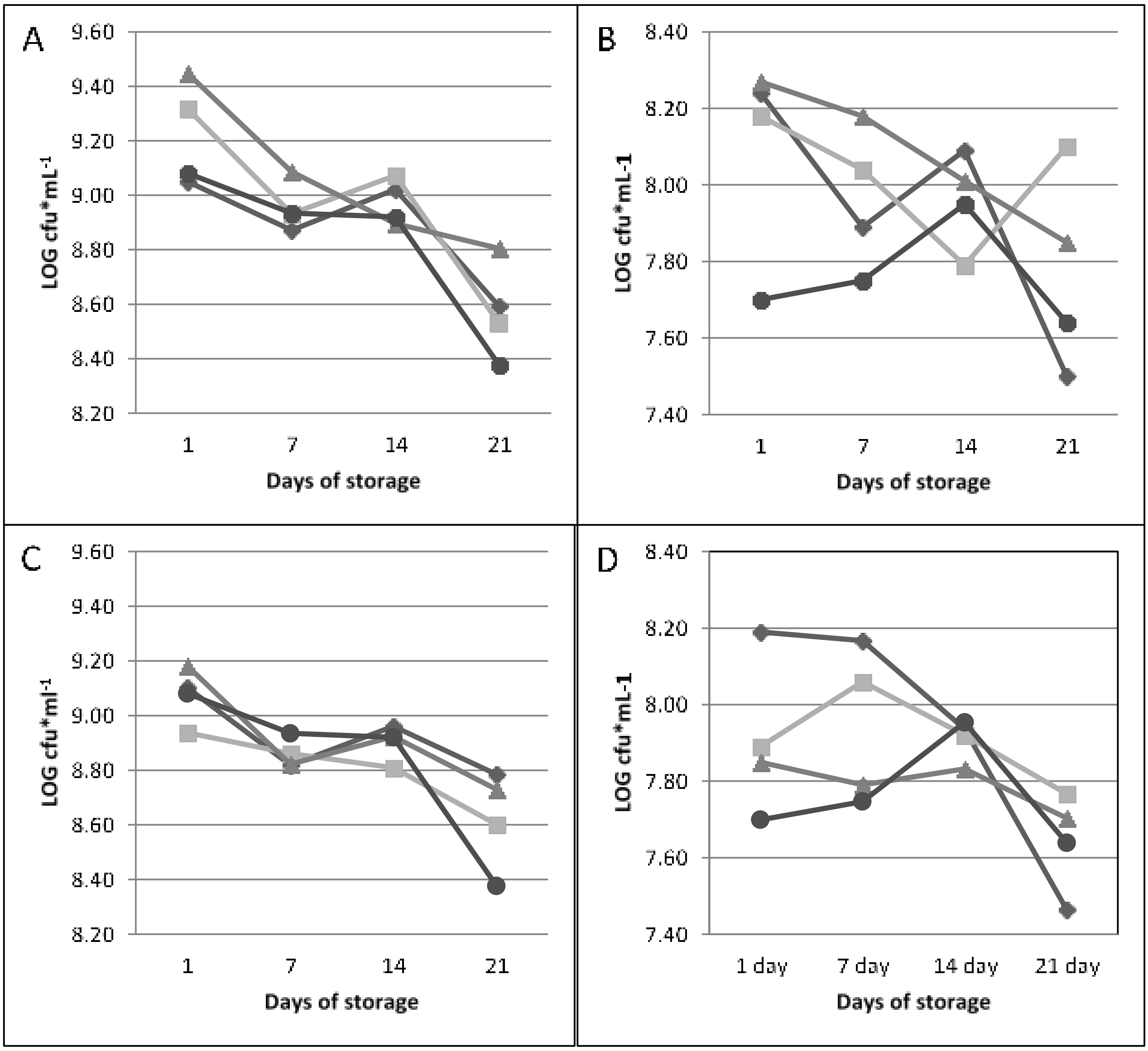
Fig. 1: HPLC-DAD chromatograms of yogurts added with 6% of hazelnut skin at 7^{th} days of storage. a) Georgia; b) Tonda Gentile Trilobata; c) San Giovanni hazelnut varieties. 1 = gallic acid; 2 = protocatechuic acid; 3 = procyanidin B1; 4 = gallocatechingallate; 5 = 3-coumaric acid; 6 = 2-coumaric acid; 7 = rutin identified compounds.

Fig. 2: *Streptococcus thermophilus* (A) and *Lactobacillus delbrueckii* subsp. *bulgaricus* (B) counts in fortified yogurts with 0% (control) and 3% of hazelnut skins during 3 weeks of storage at 4 °C. *Streptococcus thermophilus* (C) and *Lactobacillus delbrueckii* subsp. *bulgaricus* (D) counts in fortified yogurts with 0% (control) and 6% of hazelnut skins during 3 weeks of storage at 4 °C.

0% (Control) and 3% Geogia, 6% Geogia, 3% San Giovanni, 6% San Giovanni, 3% Tonda Gentile Trilobata, 6% Tonda Gentile Trilobata hazelnut varieties fortification.

Fig. 3: Linking of odour, texture, taste, flavour and acceptance expressed by 20 consumers for the control and fortified yogurts.

0% (Control) and 3% Geogia, 6% Geogia, 3% San Giovanni, 6% San Giovanni, 3% Tonda Gentile Trilobata, 6% Tonda Gentile Trilobata hazelnut varieties fortification. Histograms with different letters were significantly different at $p < 0.05$.


A
B
C
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LOG cfu*mL-1
Days of storage
1
7
14
21
1 day
7 day
14 day
21 day
9.60
9.40
9.20
9.00
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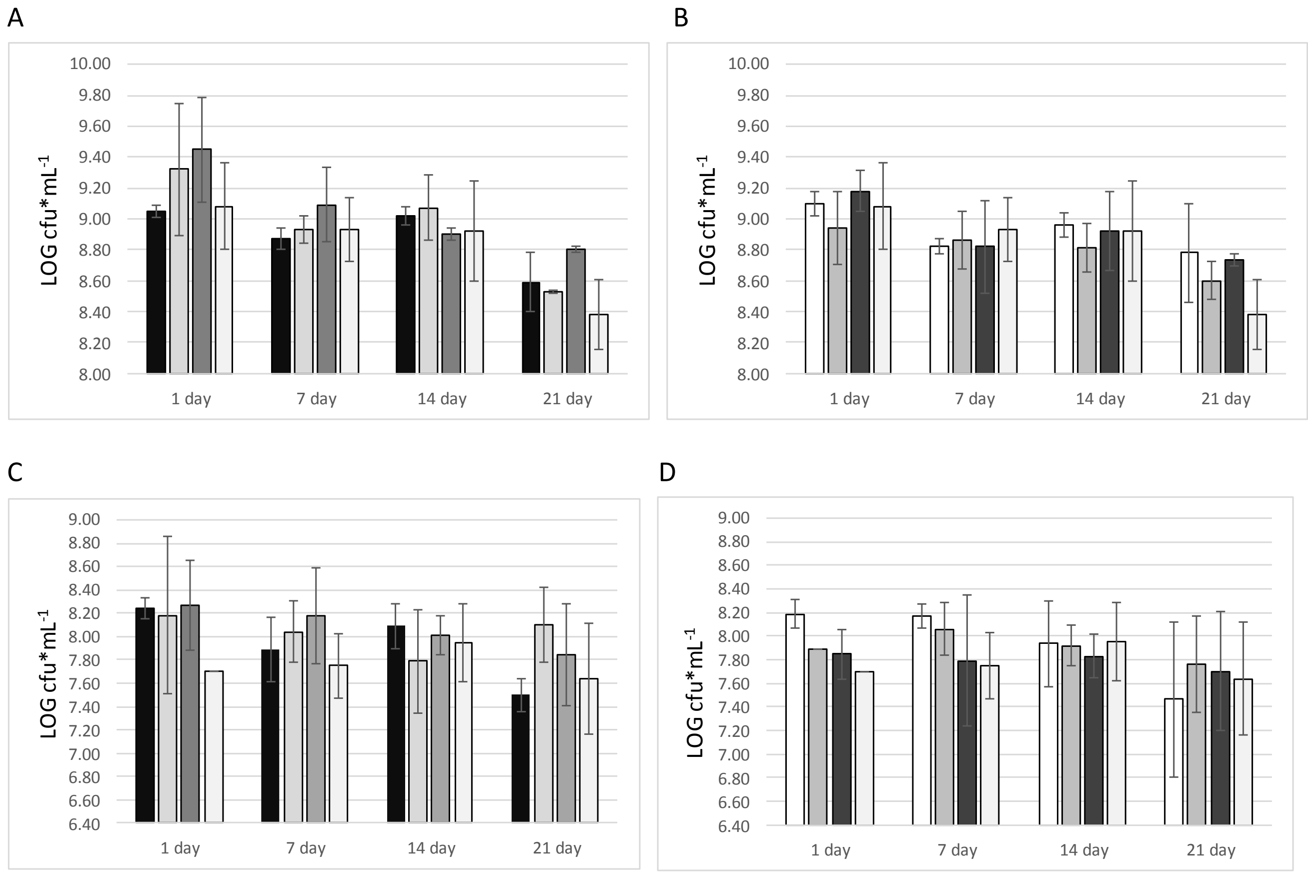
8.00
7.80
7.60
7.40


A
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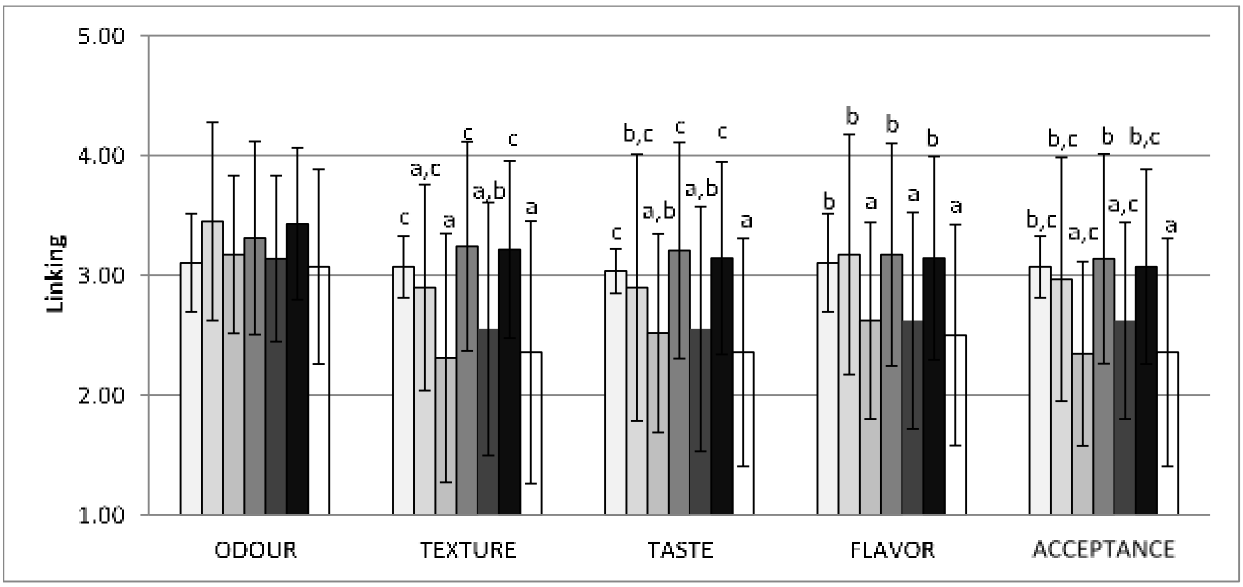
Linking
ODOUR
TEXTURE
TASTE
FLAVOR
ACCEPTANCE

Table 1: Chemical composition, total phenolic content (TPC) and DPPH radical scavenging activity (RSA) of hazelnut skin (HS)[W].

Composition	*Hazelnut varietals*			Significance
	Georgia	San Giovanni	TGT	
Humidity (g/Kg)	43.13 ± 0.15 ^{a}	60.20 ± 0.16 ^{b}	47.14 ± 0.15 ^{a}	**
Protein (g/kg dw)	93.90 ± 1.36	91.67 ± 0.83	88.46 ± 1.14	ns
Total lipid (g/kg dw)	109.86 ± 1.68 ^{a}	187.55 ± 1.45 ^{c}	171.95 ± 1.58 ^{b}	***
Carbohydrates (g/kg dw)	174.57 ± 34.28 ^{a}	183.33 ± 1.00 ^{b}	190.98 ± 2.10 ^{b}	***
Ash (g/kg dw)	21.56 ± 0.52 ^{a}	25.96 ± 0.53 ^{b}	24.66 ± 0.64 ^{b}	***
Total dietary fibre (g/kg dw)	568.44 ± 5.53 ^{b}	543.26 ± 14.57 ^{a}	542.85 ± 29.70 ^{a}	**
Soluble dietary fibre (g/kg dw)	87.57 ± 1.79 ^{c}	54.26 ± 4.60 ^{b}	45.12 ± 2.10 ^{a}	***
Insoluble dietary fibre (g/kg dw)	499.30 ± 3.48 ^{b}	464.54 ± 4.10 ^{a}	466.60 ± 4.96 ^{a,b}	***
TPC (GAE μg/g dw)	195.76 ± 4.93 ^{b}	153.29 ± 5.95 ^{a}	160.05 ± 2.84 ^{a}	***
RSA (TE μM/g dw)	1004.98 ± 21.23 ^{b}	984.66 ± 16.78 ^{b}	854.47 ± 21.59 ^{a}	***

[W] Data are expressed as mean ± SD (n = 3). Means followed by different letters were significantly different at $p < 0.05$.
Abbreviations: TGT = Tonda Gentile Trilobata, dw = dry weight; GAE = gallic acid equivalent and TE = trolox equivalent.
Significance: * p < 0.05; ** $p < 0.01$; *** $p < 0.001$; ns = not significant.

Table 2: Chemical composition of yogurts with 0% (control), 3% and 6% content in hazelnut skin (HS) [W].

Composition			*Hazelnut varietals*												
			Geogia				*San Giovanni*				*TGT*				
	0% (Control)		3% HS		6% HS		3% HS		6% HS		3% HS		6% HS		Significance
Humidity (g/Kg)	858.17 ± 0.76	c	833.72 ± 0.74	b	809.26 ± 0.71	a	834.23 ± 0.74	b	810.29 ± 0.71	a	833.84 ± 0.74	b	809.51 ± 0.72	a	***
Protein (g/kg dw)	261.00 ± 0.57	d	232.24 ± 0.51	c	210.78 ± 0.52	b	232.29 ± 0.40	c	210.75 ± 0.44	b	231.41 ± 0.53	c	209.30 ± 0.53	a	***
Total lipid (g/kg dw)	303.09 ± 23.84	c	269.75 ± 19.58	a,b,c	244.95 ± 16.38	a	283.46 ± 19.57	b,c	268.78 ± 16.35	a,b,c	280.55 ± 19.57	a,b,c	263.76 ± 16.37	a,b	***
Carbohydrates (g/kg dw)	382.90 ± 18.99	b	346.94 ± 17.75	a	320.21 ± 18.19	a	348.95 ± 15.79	a	323.58 ± 13.45	a	349.88 ± 15.84	a	325.30 ± 13.53	a	***
Ash (g/kg dw)	59.70 ± 1.93	c	53.11 ± 1.49	b	48.22 ± 1.17	a	53.96 ± 1.48	b	49.67 ± 1.15	a	53.67 ± 1.47	b	49.18 ± 1.14	a	***
Total dietary fibre (g/kg dw)	- ± -	a	98.14 ± 0.77	b	171.13 ± 1.37	d	92.41 ± 2.75	b	161.50 ± 4.89	c	93.39 ± 4.81	b	162.93 ± 8.47	c	***
Soluble dietary fibre (g/kg dw)	- ± -	a	15.12 ± 0.30	e	26.36 ± 0.52	f	9.23 ± 0.93	c	16.13 ± 1.63	e	7.76 ± 0.35	b	13.54 ± 0.61	d	***
Insoluble dietary fibre (g/kg dw)	- ± -	a	86.21 ± 0.52	c	150.31 ± 0.90	f	79.02 ± 0.70	b	138.10 ± 1.26	d	80.28 ± 0.70	b	140.06 ± 1.25	e	***

[W] Data are expressed as mean ± SD (n = 3). Means followed by different letters were significantly different at $p < 0.05$.

Abbreviations: TGT = Tonda Gentile Trilobata, dw = dry weight.

Significance: * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

Table 3: pH, acidity (express as lactic acid %) and syneresis (express as whey %) of yogurt during 3 week of storage at 4 °C[W].

				Storage period (days)											
Parameter	*Hazelnut varietals*	*HS %*		1			7			14			21		Significance
pH	Control	0	A	4.46 ± 0.02	a	B	4.38 ± 0.01	b	A	4.29 ± 0.00	c	B	4.24 ± 0.01	d	***
	Geogia	3	A	4.47 ± 0.01	d	A	4.37 ± 0.00	c	A	4.29 ± 0.01	b	B	4.24 ± 0.00	a	***
		6	A	4.46 ± 0.02	c	C	4.43 ± 0.01	c	C	4.32 ± 0.01	b	B	4.27 ± 0.01	a	***
	San Giovanni	3	A,B	4.48 ± 0.01	d	A	4.37 ± 0.01	c	A,B	4.30 ± 0.01	b	B	4.25 ± 0.01	a	***
		6	B	4.52 ± 0.03	c	C	4.43 ± 0.01	b	B	4.29 ± 0.01	a	B	4.26 ± 0.03	a	***
	TGT	3	A,B	4.48 ± 0.02	c	A	4.36 ± 0.00	b	C	4.33 ± 0.00	b	A	4.21 ± 0.01	a	***
		6	B	4.52 ± 0.03	c	D	4.45 ± 0.00	b	A	4.28 ± 0.01	a	B	4.24 ± 0.02	a	***
Significance			*			***			***			*			
Acidity	Control	0	A	0.98 ± 0.03	a	A	1.18 ± 0.03	a,b	B,C	1.40 ± 0.15	b	A,B	1.46 ± 0.20	b	***
	Geogia	3	B	1.07 ± 0.05		A	1.29 ± 0.08		A,B	1.24 ± 0.17		A,B	1.49 ± 0.09		ns
		6	C	1.14 ± 0.02		A	1.31 ± 0.10		B,C	1.41 ± 0.19		A,B	1.54 ± 0.06		ns
	San Giovanni	3	C	1.17 ± 0.00	a	A	1.17 ± 0.00	a	B,C	1.54 ± 0.15	b	A,B,C	1.68 ± 0.22	b	*
		6	C	1.14 ± 0.02		B	1.68 ± 0.43		C	1.69 ± 0.28		B,C	1.76 ± 0.01		ns
	TGT	3	C,D	1.20 ± 0.05		A	1.17 ± 0.21		A	0.99 ± 0.25		A	1.26 ± 0.51		ns
		6	D	1.25 ± 0.05	a	A	1.35 ± 0.17	a,b	B,C	1.42 ± 0.21	a,b	C	2.06 ± 0.02	b	*
Significance			***			*			*			*			
Syneresis	Control	0	A	35.34 ± 0.10	b	A	32.98 ± 0.58	a	A	31.76 ± 0.95	a	A	32.32 ± 0.10	a	*
	Geogia	3	B	40.52 ± 0.26		B	40.77 ± 1.30		B	40.41 ± 0.25		B	41.58 ± 0.60		ns
		6	D	46.73 ± 0.11	a	D	51.75 ± 0.18	c	C,D	49.93 ± 0.03	b	D	52.94 ± 0.08	d	***
	San Giovanni	3	B	40.76 ± 0.04		B	41.66 ± 0.43		B	40.72 ± 0.95		B	41.37 ± 0.31		ns
		6	E	48.24 ± 0.76		D	51.83 ± 0.49		C	48.55 ± 0.81		C	50.18 ± 1.37		ns
	TGT	3	C	43.79 ± 0.83	b	B	40.87 ± 0.07	a	B	39.87 ± 0.23	a	B	40.94 ± 0.94	a	*
		6	F	51.75 ± 0.18		C	50.30 ± 0.15		D	50.21 ± 0.90		C	51.09 ± 0.41		ns
Significance			***			***			***			***			

[W] Data are expressed as mean ± SD (n = 3).
Abbreviations: HS % = hazelnut skin content (%), TGT = Tonda Gentile Trilobata.
Means followed by different lowercase letters in same row within each concentration were significantly different at *p* < 0.05; means followed by different capital letters in same column within each storage time were significantly different at p < 0.05.
Significance: * p < 0.05; ** *p* < 0.01; *** *p* < 0.001; ns = not significant.

Table 4: Total phenolic content (TPC) and DPPH radical scavenging activity (RSA) of yogurt during 3 week of storage at 4 °C[W].

			Storage period (days)				
Parameter	*Hazenul varietals*	*HS %*	1	7	14	21	*Significance*
TPC (GAE µg/g dry matter)	Control	0	A 8.06 ± 0.28 b,c	A 7.82 ± 0.02 b	A 8.33 ± 0.07 c	A 7.23 ± 0.15 a	***
	Georgia	3	B 10.64 ± 0.61 a	B 11.51 ± 0.35 a	B 13.65 ± 0.10 b	B 13.77 ± 0.21 b	***
		6	C 15.38 ± 1.36 a	C 17.27 ± 1.38 a,b	E 20.89 ± 0.44 c	C 19.43 ± 1.84 b,c	**
	San Giovanni	3	B 10.30 ± 0.12 a	B 10.72 ± 0.59 a	B 12.71 ± 0.15 b	B 13.12 ± 0.37 b	***
		6	C 14.10 ± 0.96 a	C 16.48 ± 1.10 b	C 17.07 ± 0.55 b	C 17.86 ± 0.80 b	**
	TGT	3	B 10.67 ± 0.03 a	B 11.49 ± 0.52 a,b	B 13.56 ± 1.90 b,c	B 14.56 ± 0.16 c	**
		6	C 14.12 ± 0.47 a	C 16.42 ± 0.51 b	D 18.97 ± 0.28 c	C 18.48 ± 0.25 c	***
Significance			***	***	***	***	
RSA (TE µM/g dry matter)	Control	0	A 9.73 ± 0.41 a	A 8.89 ± 0.32 a	A 10,00 ± 0.18 a	A 12.02 ± 1.09 b	**
	Georgia	3	B 19.50 ± 0.78 a	B 20.15 ± 0.33 a	B 24.67 ± 0.51 a,b	B,C 29.71 ± 3.97 b	***
		6	C,D 29.40 ± 2.75 a	C 31.80 ± 2.22 a,b	F 39.16 ± 1.17 c	D 38.41 ± 3.76 b,c	**
	San Giovanni	3	B 17.84 ± 1.20 a	B 18.95 ± 0.97 a	B 23.22 ± 0.10 b	B 25.27 ± 1.66 b	***
		6	D 25.44 ± 2.28 a	C 29.49 ± 2.33 a,b	D 31.71 ± 1.28 b,c	C,D 35.49 ± 1.08 c	***
	TGT	3	B 20.01 ± 0.14 a	B 21.71 ± 0.91 a	C 28.35 ± 0.61 b	C,D 33.89 ± 2.30 c	***
		6	C 27.24 ± 1.85 a	C 31.26 ± 0.92 a,b	E 35.48 ± 0.45 b	E 47.29 ± 3.00 c	***
Significance			***	***	***	***	

[W] Data are expressed as mean ± SD (n = 3).
Abbreviations: HS % = hazelnut skin content (%), TGT = Tonda Gentile Trilobata, GAE = Gallic acid equivalent, TE = Trolox equivalent.
Means followed by different lowercase letters in same row within each concentration were significantly different at $p < 0.05$; means followed by different capital letters in same column within each storage time were significantly different at p < 0.05.
Significance: * p < 0.05; ** $p < 0.01$; *** $p < 0.001$.

Table 5: Phenolic compound concentration (mg/kg) of yogurt during 3 week of storage at 4 °C[W].

				Storage period (days)											
Parameter	*Hazelnut varietals*	*HS %*		1			7			14			21		Significance
Gallic acid	Geogia	3	A	4.21 ± 0.91		A	5.89 ± 0.31		A	5.89 ± 0.10		A	7.22 ± 1.71		ns
		6	B,C	10.62 ± 2.01		B	14.02 ± 0.81		B	14.61 ± 0.11		C	15.19 ± 0.91		ns
	San Giovanni	3	A,B	6.11 ± 0.32	a	A	7.41 ± 0.50	a,b	A	7.10 ± 0.42	a,b	A,B	8.32 ± 0.21	b	*
		6	B	9.51 ± 0.41	a	C	17.42 ± 1.40	a,b	B	12.61 ± 2.91	a,b	C	16.71 ± 1.61	b	*
	TGT	3	A,B	8.33 ± 1.01	a	B	10.71 ± 0.92	a,b	B	12.02 ± 0.12	a,b	B,C	13.14 ± 1.60	b	*
		6	C	15.53 ± 1.71	a	D	22.53 ± 0.60	a	C	20.81 ± 0.22	a	D	26.71 ± 1.60	b	**
Significance			**			***			***			***			
Protocatechuic acid	Geogia	3	B	15.21 ± 1.20		B	18.71 ± 0.70		C	20.11 ± 0.40		B	23.31 ± 4.41		ns
		6	C	30.71 ± 5.81		C	38.82 ± 2.92		D	42.89 ± 0.60		C	43.12 ± 0.70		ns
	San Giovanni	3	A	4.61 ± 0.22		A	5.61 ± 0.22		A	5.73 ± 0.60		A	6.60 ± 1.10		ns
		6	A,B	8.51 ± 0.00		A	10.91 ± 0.60		B	11.04 ± 2.01		A	12.52 ± 0.91		ns
	TGT	3	A,B	9.50 ± 1.80		A	11.42 ± 1.91		B	14.51 ± 0.10		A	14.44 ± 1.71		ns
		6	B	15.41 ± 0.10	a	B	22.73 ± 0.40	b	C	22.52 ± 0.61	b	B	28.01 ± 1.61	c	**
Significance			**			***			***			***			
Procyanidin B1	Geogia	3	A,B	40.31 ± 4.70		C	47.71 ± 2.21		C	45.71 ± 0.30		B	47.74 ± 9.83		ns
		6	B	63.82 ± 17.71		D	70.10 ± 5.01		D	70.20 ± 1.80		C	66.72 ± 2.01		ns
	San Giovanni	3	A	17.11 ± 0.51		A	19.54 ± 1.32		A	16.83 ± 0.61		A	18.33 ± 1.21		ns
		6	A	25.11 ± 4.12		B	32.12 ± 1.81		A,B	25.31 ± 1.50		A	26.01 ± 1.61		ns
	TGT	3	A	30.90 ± 2.81		B	33.33 ± 3.72		B,C	35.04 ± 0.00		A	28.04 ± 1.93		ns
		6	A,B	44.01 ± 2.60		C,D	58.50 ± 2.50		C	46.91 ± 10.20		C	66.32 ± 1.80		ns
Significance			**			***			***			***			
Gallocatechingallate	Geogia	3		4.10 ± 0.30		A	3.93 ± 0.11			3.71 ± 0.00			3.51 ± 0.00		ns
		6		4.72 ± 0.11		A,B	4.50 ± 0.00			4.42 ± 0.71			4.02 ± 0.00		ns
	San Giovanni	3		4.73 ± 0.00		B	4.84 ± 0.23			4.54 ± 0.21			3.84 ± 0.52		ns
		6		5.01 ± 0.42		B	5.02 ± 0.00			4.51 ± 0.40			4.11 ± 0.00		ns
	TGT	3		4.83 ± 0.21		B	4.82 ± 0.11			5.63 ± 1.40			4.22 ± 0.31		ns
		6		4.89 ± 0.21		B	5.01 ± 0.30			4.52 ± 0.21			4.62 ± 0.31		ns
Significance			NS			**			NS			NS			
3-Coumaric acid	Geogia	3		0.10 ± 0.00			1.90 ± 0.00			0.17 ± 0.00			0.19 ± 0.00		ns
		6		0.10 ± 0.00			1.90 ± 0.00			0.18 ± 0.00			0.10 ± 0.00		ns
	San Giovanni	3		0.19 ± 0.00			1.80 ± 0.00			0.19 ± 0.00			0.17 ± 0.00		ns
		6		0.22 ± 0.00			3.00 ± 0.00			0.10 ± 0.00			0.22 ± 0.11		ns
	TGT	3		0.10 ± 0.00			2.00 ± 0.00			0.20 ± 0.00			0.29 ± 0.00		ns
		6		0.21 ± 0.00			1,00 ± 0.00			0.1 ± 0.00			0.1 ± 0.00		ns
Significance			NS			NS			NS			NS			
2-Coumaric acid	Geogia	3		< LOQ			< LOQ			< LOQ			< LOQ		
		6		< LOQ			< LOQ			< LOQ			< LOQ		
	San Giovanni	3		ND			ND			ND			< LOQ		
		6		< LOQ			ND			ND			ND		
	TGT	3		ND			ND			ND			ND		
		6		ND			ND			ND			ND		
Significance															
Rutin	Geogia	3	A	0.10 ± 0.00		A	0.10 ± 0.00		A,B	0.29 ± 0.00		A	0.39 ± 0.00		ns
		6	B	0.80 ± 0.10		B,C	0.89 ± 0.00		B	0.71 ± 0.10		A,B	0.61 ± 0.10		ns
	San Giovanni	3		< LOQ		A	0.10 ± 0.00	a	A	0.10 ± 0.00	a	A	0.32 ± 0.00	b	*
		6	A,B	0.51 ± 0.20		C	1.22 ± 0.22		A,B	0.59 ± 0.30		C	1.21 ± 0.20		ns
	TGT	3		< LOQ		A	0.10 ± 0.00		A	0.11 ± 0.00		A	0.31 ± 0.00		ns
		6	A,B	0.31 ± 0.10	a	B	0.51 ± 0.11	a	A,B	0.51 ± 0.10	a	B,C	1.11 ± 0.20	b	*
Significance			**			***			*			**			

[W] Data are expressed as mean ± SD (n = 3).
Abbreviations: HS % = hazelnut skin content (%), TGT = Tonda Gentile Trilobata, LOQ = limit of quantification.
Means followed by different lowercase letters in same row within each concentration were significantly different at $p < 0.05$; means followed by different capital letters in same column within each storage time were significantly different at $p < 0.05$.
Significance: * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; ns or NS = not significant.

Table 6: Sugar and organic acid concentrations (g/kg) of yogurt during 3 week of storage at 4 °C[W].

				Storage period (days)											
Parameter	Hazelnul varietals	HS %		1			7			14			21		Significance
Lactose	Control	0		48.9 ± 0.04	b		47.05 ± 0.19	a		46.24 ± 0.39	a		45.83 ± 0.89	a	*
	Geogia	3		48.02 ± 0.27	b		47.62 ± 2.00	b		45.17 ± 0.48	a,b		44.05 ± 0.11	a	*
		6		47.28 ± 0.73	b		45.76 ± 0.05	a,b		44.70 ± 0.32	a		44.11 ± 1.12	a	*
	San Giovanni	3		47.90 ± 0.30	c		46.32 ± 0.37	b		45.12 ± 0.33	a		44.80 ± 0.28	a	**
		6		46.29 ± 1.05			46.14 ± 0.73			44.70 ± 0.24			44.91 ± 2.48		ns
	TGT	3		49.52 ± 2.69			46.81 ± 1.10			45.44 ± 0.35			44.77 ± 0.07		ns
		6		46.89 ± 0.10			46.37 ± 0.67			45.32 ± 0.18			46.29 ± 1.59		ns
Significance			NS			NS			NS			NS			
Glucose	Control	0	A,B,C	0.37 ± 0.04	a	B	0.37 ± 0.04	b	C	0.92 ± 0.00	c	B	0.96 ± 0.00	c	***
	Geogia	3	A,B	0.35 ± 0.05	a	B	0.69 ± 0.04	b	B,C	0.81 ± 0.03	b	A	0.23 ± 0.06	a	***
		6	B,C	0.40 ± 0.04	b	C	0.87 ± 0.00	d	A	0.49 ± 0.01	c	A	0.17 ± 0.02	a	***
	San Giovanni	3	A	0.31 ± 0.02	a	A,B	0.63 ± 0.05	b	B,C	0.81 ± 0.09	c	A	0.33 ± 0.06	a	**
		6	C	0.43 ± 0.01		C	0.82 ± 0.00		B	0.67 ± 0.01		A	0.53 ± 0.41		ns
	TGT	3	A	0.31 ± 0.00	a	A	0.59 ± 0.01	b	B,C	0.80 ± 0.13	c	A	0.23 ± 0.01	a	*
		6	C	0.43 ± 0.02	b	C	0.83 ± 0.67	c	A	0.37 ± 0.07	b	A	0.21 ± 0.01	a	***
Significance			*			***			***			*			
Galactose	Control	0	C	11.97 ± 0.24	a	D	11.97 ± 0.24	a,b	C	12.91 ± 0.11	b,c	C	13.33 ± 0.29	c	*
	Geogia	3	B,C	11.46 ± 0.20	b	C,D	12.27 ± 0.20	b	A	11.09 ± 0.03	b	A,B	9.42 ± 1.15	a	*
		6	A,B	10.90 ± 0.10	b	A,B	11.25 ± 0.05	b,c	A	11.44 ± 0.24	c	A	8.41 ± 0.05	a	***
	San Giovanni	3	B,C	11.51 ± 0.30	a,b	C	12.09 ± 0.02	b	B,C	12.42 ± 0.13	b	B	10.17 ± 0.96	a	*
		6	A,B	10.89 ± 0.01		B	11.47 ± 0.04		B	12.18 ± 0.02		B	10.81 ± 0.98		ns
	TGT	3	B,C	11.63 ± 0.78	b	C	12.07 ± 0.11	b	A	11.31 ± 0.71	b	A,B	9.48 ± 0.20	a	*
		6	A	10.49 ± 0.22	b	A	11.13 ± 0.01	c	A	11.23 ± 0.01	c	A	7.97 ± 0.12	a	***
Significance			*			***			**			*			
Pyruvic acid	Control	0	B	0.89 ± 0.00		C	0.89 ± 0.00			0.91 ± 0.00			0.91 ± 0.02		ns
	Geogia	3	B	0.88 ± 0.02		C	0.91 ± 0.02			0.87 ± 0.01			0.89 ± 0.02		ns
		6	B	0.87 ± 0.00	a	B	0.87 ± 0.00	a,b		0.88 ± 0.01	b		0.90 ± 0.00	c	*
	San Giovanni	3	B	0.86 ± 0.01	a	B,C	0.89 ± 0.00	b,c		0.88 ± 0.00	a,b		0.90 ± 0.00	c	**
		6	A	0.81 ± 0.01		A	0.85 ± 0.00			0.86 ± 0.00			0.86 ± 0.02		ns
	TGT	3	B	0.88 ± 0.04		B,C	0.89 ± 0.01			0.86 ± 0.03			0.89 ± 0.01		ns
		6	A	0.79 ± 0.00	a	A	0.84 ± 0.00	b		0.85 ± 0.01	b		0.92 ± 0.02	c	**
Significance			**			***			NS			NS			
Lactic acid	Control	0	C	18.15 ± 0.44	a	D	18.15 ± 0.44	a,b	D	19.52 ± 0.18	b,c	C	20.39 ± 0.53	c	*
	Geogia	3	B,C	17.38 ± 0.46	a	C,D	18.43 ± 0.34	b	A,B,C	18.21 ± 0.19	a,b	B	18.98 ± 0.13	b	*
		6	A,B	16.31 ± 0.32	a	A,B	16.37 ± 0.13	a,b	A,B	17.78 ± 0.38	b	A,B	18.5 ± 0.07	b	**
	San Giovanni	3	B,C	17.51 ± 0.63	a	C,D	18.29 ± 0.04	a,b	C	18.68 ± 0.03	b,c	B,C	19.4 ± 0.20	c	*
		6	A,B	16.32 ± 0.19	a	B	16.76 ± 0.01	a,b	B,C	18.36 ± 0.04	a,b	A	17.56 ± 1.13	b	*
	TGT	3	B,C	17.61 ± 1.17	a	C	18.23 ± 0.21	a	A	17.72 ± 0.47	b	B,C	19.37 ± 0.06	c	*
		6	A	15.61 ± 0.51	a	A	16.11 ± 0.00	a	A,B,C	18.18 ± 0.00	b	B	19.06 ± 0.22	c	***
Significance			*			***			**			*			
Malic acid	Control	0	A	- ± -		A	- ± -		A	- ± -		A	- ± -		
	Geogia	3	B	0.08 ± 0.01		C	0.07 ± 0.01		B	0.08 ± 0,00		B	0.08 ± 0.00		ns
		6	B	0.07 ± 0.00		B	0.05 ± 0.00		C	0.10 ± 0.01		B	0.07 ± 0.00		ns
	San Giovanni	3	C	0.17 ± 0.01		E	0.17 ± 0.00		D	0.16 ± 0.00		C	0.16 ± 0.00		ns
		6	D	0.40 ± 0.05		F	0.33 ± 0.01		E	0.32 ± 0.01		C	0.33 ± 0.01		ns
	TGT	3	B	0.07 ± 0.00		B	0.07 ± 0.01		B	0.07 ± 0.01		B	0.07 ± 0.00		ns
		6	C	0.21 ± 0.00		D	0.15 ± 0.00		D	0.15 ± 0.00		B	0.12 ± 0.05		ns
Significance			***			***			***			***			
Citric acid	Control	0		2.72 ± 0.01			2.72 ± 0.01			2.74 ± 0.01			2.77 ± 0.07		ns
	Geogia	3		2.67 ± 0.02			2.75 ± 0.08			2.68 ± 0.01			2.68 ± 0.03		ns
		6		2.64 ± 0.00			2.63 ± 0.02			2.66 ± 0.03			2.70 ± 0.01		ns
	San Giovanni	3		2.72 ± 0.02			2.73 ± 0.00			2.73 ± 0.01			2.68 ± 0.11		ns
		6		2.71 ± 0.03			2.74 ± 0.01			2.75 ± 0.03			2.75 ± 0.00		ns
	TGT	3		2.76 ± 0.14			2.71 ± 0.03			2.70 ± 0.02			2.73 ± 0.01		ns
		6		2.66 ± 0.00			2.67 ± 0.01			2.71 ± 0.02			2.85 ± 0.08		ns
Significance			NS			NS			NS			NS			

[W] Data are expressed as mean ± SD (n = 3).

Abbreviations: HS % = hazelnut skin content (%), TGT = Tonda Gentile Trilobata.

Means followed by different lowercase letters in same row within each concentration were significantly different at $p < 0.05$; means followed by different capital letters in same column within each storage time were significantly different at $p < 0.05$.

Significance: * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; ns or NS = not significant.