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STORAGE STABILITY OF LOW AND HIGH HEAT TREATED HAZELNUT BEVERAGES

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ABSTRACT

In this study, the changes in the quality parameters of hazelnut beverages treated with low (LHT; 72°C for 20 min) and high temperature (HHT; 105°C for 1 min) after high pressure homogenization process were determined during short (10 days) and long term (120 days) storage periods, respectively. Microbial viability was not detected in any thermally treated samples. Although pH and titration acidity values of LHT samples did not show important change during the storage, the pH values of HHT samples decreased significantly. While protein solubility of LHT samples increased during storage, it slightly decreased in HHT samples. Rheological properties and also serume stability of LHT and HHT hazelnut samples changed during storage. Hydroperoxide index value slightly increased in LHT samples while it increased more than three times in HHT samples during storage. As a result, the LHT and HHT hazelnut beverages had different structural and physicochemical properties during storage.

Keywords: Hazelnut beverage, heat treatment, storage stability, hydroperoxide index

DÜŞÜK VE YÜKSEK ISIL İŞLEM UYGULANMIŞ FINDIK İÇECEKLERİNİN DEPOLAMA STABİLİTELERİ

ÖZ

Bu çalışmada, yüksek basınç homojenizasyonu işleminden sonra düşük (LHT; 72 °C'de 20 dak) ve yüksek (HHT; 105 °C'de 1 dak) ısıl işlem uygulanan fındık içeceklerinin sırası ile kısa (10 gün) ve uzun süreli (120 gün) depolama süresince kalite parametrelerindeki değişimler belirlenmiştir. Isıl işlem görmüş içeceklerde canlı bakteri tespit edilmemiştir. Düşük sıcaklıkta ısıl işlem görmüş örneklerin pH ve titrasyon asitliği değerleri depolama boyunca değişmezken yüksek sıcaklıkta ısıl işlem görmüş örneklerin pH değerlerinde azalma gözlenmiştir. LHT örneklerinin protein çözünürlükleri depolama boyunca artarken HHT örneklerinde azalmıştır. LHT ve HHT fındık içeceklerinin reolojik özellikleri ve serum stabiliteleri depolama süresince değişiklik göstermiştir. Depolama boyunca LHT örneklerinde hidroperoksit indeks değeri hafifçe artarken HHT örneklerinde bu artış üç kat daha fazla olmuştur. Sonuç olarak, LHT ve HHT örneklerinde depolama boyunca farklı yapısal ve fizikokimyasal değişimler meydana geldiği gözlenmiştir.

Anahtar kelimeler: Fındık içeceği, ısıl işlem, depolama stabilitesi, hidroperoksit indeks

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INTRODUCTION

Vegetable-based beverages represent a leading growth trend in consumer packaged foods sector. People who are vegetarians, flexitarian, vegan or suffer from lactose intolerance and milk allergy want an alternative to animal milks. Due to the increased trend, different vegetable-based beverages especially soy, almond, cashew and coconut beverages commonly put on the markets and also hazelnut beverage from hazelnut (Bernat et al., 2015) or cold pressed hazelnut cakes which is a by-product from hazelnut oil production (Gul et al., 2017, Gul et al., 2018a, Gul et al., 2018b) has been spread to all over the world. According to the study reported by Şimşek and Aslantaş (1999)and Alasalvar et al. (2003), hazelnut plays an important role in human nutrition because of its special composition of fat, protein, carbohydrate, dietary fiber, vitamins, minerals, phytosterols, and antioxidant phenolics. Its major health benefits are help to reduce LDL cholesterol and increase HDL cholesterol. It is also filled with antioxidants that wipe out damaging free radicals (Oliveira et al., 2008). Hazelnut is evaluated in food industry as the main ingredient (roasted, whitened, sliced, etc.) or raw material for chocolate and oil production. As a higher protein source, hazelnut cake can be evaluated for human nutrition in the production of hazelnut beverage and products.

Vegetable-based beverages are emulsified products that product stability needs to be modified with homogenization and thermal treatment processes. These processes ensure high-quality food products which meaning as guaranty of good nutritional quality, long shelf life, and high colloidal stability. In our previous studies the optimum production conditions and heat treatment parameters were determined according to good stability, physical properties and microbiological safety (Gul et al., 2018b). According to stability and microbiological results, 100 MPa were selected as homogenization pressure, and 72 °C for 20 min (low heat treatment) and 105 °C 1 min (high heat treatment) were chosen as thermal treatment parameters. In the literature there are some studies about the effect of homogenization and heat treatment on the vegetable beverages (Bernat et al., 2015, Briviba et al., 2016, Rosello-Soto et al., 2018). However, there is no study that has systematically evaluated the changes that occurring in homogenized hazelnut beverage during storage. The present study was conducted to investigate the variation of microbiological, physicochemical and rheological properties of low (at 72 °C for 20 min) and high (at 105 °C for 1 min) thermally treated hazelnut beverages during storage period.

MATERIAL AND METHODS

Material and preparation of hazelnut beverages

The cold press hazelnut cake (4.78% moisture, 95.22% dry matter, 48.23% protein in dry matter, 31.42% carbohydrate in dry matter, 9.78% lipid in dry matter, and 5.79% ash in dry matter), was obtained after cold press extraction of hazelnut oil from whitened hazelnut (Gursoy Hazelnut Production Factory, Ordu, Turkey) and used for hazelnut beverage production. Hazelnut cakes were grounded by using a blender (Waring laboratory blender, Conair Corporation, Stamford, CT, USA) for 10 min and the grounded hazelnuts were mixed with distilled water at 1:10 w/v ground nut-water ratio. The hazelnut-water mixture was homogenized with the homogenizer at 10000 rpm for 10 min (IKA-Werke GmbH & Co. KG, Staufen, Germany). After mixing, high pressure treatment was applied at 100 MPa with two-stage homogenizer (GEA Niro Soavi – Panda PLUS 2000 Homogenizer, GEA Niro Soavi S.P.A., Parma, Italy) and samples were collected in 250 mL glass bottles for thermal treatment.

To obtain thermally treated hazelnut beverage, samples were heated with a water bath (Nuve, Ankara, Turkey) at 72 °C for 20 min for low temperature long time and heated with autoclave (Nuve, Ankara, Turkey) at 105 °C for 1 min for high temperature short time. All samples were rapidly cooled in an ice bath to a temperature of ~4 °C. The low and high heat treated samples were stored at refrigeration temperature for 10 and 120 days, respectively.

Microbiological analysis
Microbiological quality of hazelnut beverage was assessed by enumerating the following microorganisms: mesophilic aerobic bacteria were grown on Plate Count Agar (PCA, Merck, Germany) at 30 °C for 48 h and yeast-mold were grown on yeast glucose chloramphenicol agar (YGC, Merck, Darmstadt, Germany) at 25 °C for 5 day.

Physicochemical analysis
Total solids were determined gravimetrically by using an oven at 105 °C until a constant weight was obtained. Total soluble solids (°Brix) of hazelnut beverage samples were measured at 20 °C using a refractometer. The pH values of the samples were measured with a calibrated pH meter at 25 °C (Eutech Cyberscan pH 2700, Ayer Rajah Crescent, Singapore).

To perform protein solubility, the sample (1 mL) was mixed with 1 mL of Biuret reagent and homogenized by vortex for 1 min. Samples were held 20 min at room conditions, and then, the absorbance of samples was measured at 550 nm by UV spectrometer (Helios Gama, England). Protein solubility of hazelnut beverage was calculated from a standard curve of Bovine Serum Albumin (BSA) (Robinson and Hogden, 1940).

Serum separation of LHT and HHT hazelnut beverage samples was determined by the centrifuge separation method described by Valencia-Flores et al. (2013) with slight modifications. Ten g of hazelnut beverage samples were weighted in 50 mL centrifugation tubes. The samples were centrifuged (Nuve, Ankara, Turkey) at 3400 x *g* for 15 min at 10 °C. The separated part was weighed, divided by the initial weight of samples and expressed as the percentage.

Hydroperoxide Index
Hazelnut beverage (2 mL) was mixed with methanol (2 mL) and chloroform (4mL) and shaken for 30 s. The mixture was centrifuged (8000 x *g*, 20 min, 20 °C) and 1 mL of the chloroform phase was transferred to a test tube and mixed with 1 mL of Fe (II)/thiocyanate in methanol/chloroform (1:1). After 10 min reaction time, the absorbance was measured at 500 nm by UV spectrometer (Helios Gama, England). Data were expressed as meq peroxide L^{-1} of sample (Valencia-Flores et al., 2013).

Rheological measurements
Rheological properties of hazelnut beverages were measured by using Haake Mars III rheometer (Thermo Scientific, Germany) with a cone and plate system (35 mm diameter, 0.105 mm gap, 2° cone angle) (Gul et al., 2017). Temperature control was achieved with a circulator water bath at 25 °C. Flow behavior properties of hazelnut beverages were determined by recording shear stress values when shearing the samples at linearly increasing shear rates from 1 to 100 s^{-1} through 120 s. The relationship between shear stress and shear rate was described by Ostwald-de-Waele model (Eq.1).

$$\eta_{app} = K \times \dot{\gamma}^{n-1} \quad \text{(Eq.1)}$$

Where η_{app} is the apparent viscosity (Pa.s); *K* is the consistency index (Pa.s^n); $\dot{\gamma}$ is the shear rate (s^{-1}) and *n* is the flow behavior index (dimensionless). Rheowin 4 Data Manager software (version 4.20, Haake Company, Darmstadt, Germany) was used for calculations. All the rheological parameters were the mean of two measurements per duplicates of hazelnut beverage samples.

Color Properties
Color measurements were performed to determine *L** (lightness), *a** (red-green) and *b** (yellow-blue) values of the hazelnut beverage samples using a colorimeter (Minolta Chroma Meter, CR-400, Osaka, Japan). The total color difference (ΔE) (Eq. 2) was calculated by following equations;

$$Color\ difference = \Delta E = \sqrt{(L_0 - L)^2 + (a_0 - a)^2 + (b_0 - b)^2} \quad \text{(Eq. 2)}$$

where L_0, a_0, and b_0 are the initial color attributes for thermally treated hazelnut beverage samples.

Statistical analysis
Statistical analysis of the samples was performed by the SPSS statistics version 21.0 (SPSS, Chicago,

Illinois, USA). All the experiments were performed in triplicate and the results were expressed as mean ± standard deviation (SD). Differences between the samples were determined by using one-way analysis of variance (ANOVA) and multiple comparisons were performed by Duncan's test with a confidence level of 95% ($P<0.05$).

RESULTS AND DISCUSSION

Microbiological quality

The shelf life of foods determines by microbial activity and also biochemical changes occurred during storage period (Achouri et al., 2007). A fast microbial growth takes place in vegetable beverages like hazelnut and tiger nuts due to their chemical composition and neutral pH (Gul et al., 2018b, Rosello-Soto et al., 2018). So that, the preservation treatments like thermal treatment should be applied to extend of vegetable beverage quality over time (Codina-Torrella et al., 2018). The total aerobic bacteria and yeast and mould were not detected in any thermally treated hazelnut sample (below the limit of detection; <10 organism/g of sample) and also any microbiological growth could not detected during storage period. Similarly, Achouri et al. (2007) stated that all thermal treated freshly soy beverage samples contained very low numbers of microorganisms (<10 organisms/g of sample). Ukwuru and Agbodo (2011) also found that there was no growth of microorganisms on the fresh tiger nut beverage samples heated at 75 °C for 15 min (pasteurized) and 145 °C for 15 sec (ultra high temperature) and samples were microbiologically stable during storage period (4 °C) as 2 weeks for pasteurized and 6 weeks for ultra high temperature tiger nut beverages.

Physicochemical properties

The physicochemical changes of low and high thermal treated samples are given in Table 1. The pH value of the LHT samples was stable during storage period, but in HHT samples pH values decreased approximately 0.1 units during storage period of 120 d. This decrease in pH may result from chemical interactions such as lipolysis and proteolysis occurring in hazelnut beverage, which may be an indicator of product acidity (Achouri et al., 2007). The total solid contents of hazelnut beverage samples were determined between 9.42% and 9.53% at the beginning of storage. Although there was a slight increase in total solid contents of low thermal treated samples ($P<0.05$), no significant change was observed in high thermal treated samples during storage ($P>0.05$).

Table 1. The physicochemical changes of LHT and HHT hazelnut beverages during short and long time storage periods, respectively

Sample	Storage (days)	pH	Total solid (%)	Water soluble matter (°Brix)	Protein Solubility (%)	Serum separation (%)
LHT	0	6.56±0.01^{a}	9.42±0.04^{b}	4.9±0.04^{b}	3.29±0.12^{c}	49.57±0.53^{b}
	2	6.55±0.01^{a}	9.43±0.06^{b}	4.9±0.32^{b}	3.33±0.13^{c}	50.7±0.27^{b}
	4	6.55±0.02^{a}	9.45±0.01^{b}	5.13±0.05^{ab}	3.57±0.11^{b}	50.08±0.17^{b}
	6	6.53±0.02^{a}	9.47±0.04^{b}	5.22±0.05^{ab}	3.81±0.12^{a}	52.09±0.62^{ab}
	8	6.54±0.01^{a}	9.52±0.02^{a}	5.28±0.06^{ab}	3.84±0.06^{a}	53.71±0.43^{a}
	10	6.54±0.02^{a}	9.53±0.01^{a}	5.63±0.05^{a}	3.78±0.15^{a}	54.72±0.81^{a}
HHT	0	6.58±0.02^{a}	9.51±0.06^{a}	4.39±0.18^{a}	3.21±0.08^{a}	46.09±2.38^{a}
	15	6.56±0.01^{a}	9.49±0.06^{a}	4.3±0.2^{a}	3.18±0.06^{a}	43.29±1.96^{a}
	30	6.55±0.02^{a}	9.48±0.04^{a}	4.22±0.06^{a}	3.16±0.05^{a}	43±1.3^{ab}
	45	6.51±0.02^{b}	9.48±0.05^{a}	4.28±0.25^{a}	3.15±0.25^{a}	45.31±1.15^{a}
	60	6.5±0.02^{b}	9.48±0.11^{a}	3.93±0.08^{b}	3.12±0.32^{a}	45.52±0.89^{a}
	90	6.45±0.02^{b}	9.48±0.1^{a}	3.85±0.01^{b}	3.09±0.22^{a}	44.86±0.94^{a}
	120	6.47±0.01^{b}	9.49±0.03^{a}	3.97±0.03^{b}	3.1±0.16^{a}	39.17±0.25^{b}

Values are means ± Standard Deviation. $^{a-f}$ Means within the same column with different letters are significantly different at $p< 0.05$.

The °Brix value of LHT hazelnut beverage samples increased from 4.9° to 5.63° during storage, however, a slight decrease was determined in HHT hazelnut beverage samples. The increments of °Brix value of LHT samples may be due to an increase of soluble protein during storage period. The decrease of °Brix value of HHT samples can come forward with decreasing protein solubility during storage. The intensity of thermal treatment led to different aggregation behavior in the proteins during the storage period.

The serum separation value of LHT hazelnut beverage slightly increased from 49.57% to 54.72% during the storage period (Table 1). Similarly, Shimoyamada et al. (2008) reported that thermal treatments at 70 and 80 °C caused the increase in precipitation of soy beverage. The opposite situation was observed for HHT samples during the long time storage period. The serum separation value decreased from 46.09% to 39.16% in HHT samples. This could be attributed to aggregation of proteins caused to improvement on the water holding capacity of samples due to high heat treatment. Similar results found by Shimoyamada et al. (2008), who reported that soy beverage heated at higher than 90 °C showed decrease in precipitation or increase in dispersion stability. Bernat et al. (2015) were found that submitted homogenized hazelnut beverage samples to thermal treatment contribute to stabilizing the emulsions by denaturation of proteins and mainly due to a thickening effect.

Hydroperoxide Index

Hazelnut beverages contain polyunsaturated fatty acids which are very susceptible to oxidation. Oxygen, light, enzymes, and temperature are the main factors that affect the lipid oxidation. In this study the oxidation was evaluated by measuring the hydroperoxides concentration. High heat treatment at 105 °C for 1 min caused to lowering the peroxide index value (0.058 meq peroxide L^{-1}) compared to low heat treatment at 72 °C for 20 min (0.089 meq peroxide L^{-1}) (Figure 1). Similarly, Poliseli-Scopel et al. (2012) reported that the hydroperoxide value of soy beverage thermal treated at 95 °C for 30 s was higher than the other sample thermal treated at 142 °C for 6 s. However, Valencia-Flores et al. (2013) did not observe any differences between the treatments at the same homogenization pressure with different temperatures (at 55-75 °C) in the hydroperoxide index value of almonds beverages.

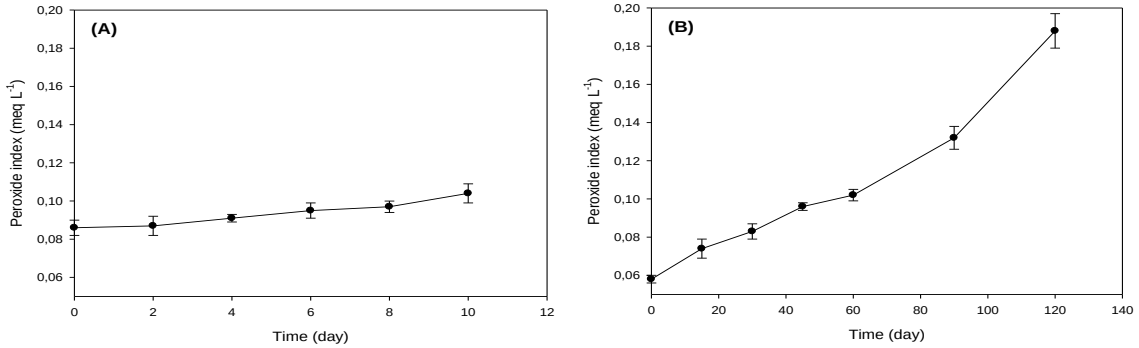

Figure 1. The peroxide index (meq L^{-1}) of LHT (a) and HHT (b) hazelnut beverages during short and long time storage periods, respectively

The hydroperoxide index of LHT samples did not significantly change during short time storage ($P>0.05$). However, the hydroperoxide index of HHT samples significantly increased during long time storage period and reached 0.188 meq peroxide L^{-1} at the end of storage period (120 days). The increment of hydroperoxide index demonstrated that autoxidation still continues to occur due to free radicals (peroxides, hydroperoxides) formed by reaction of free fatty acids with the oxygen present in air during storage period.

Rheological measurements

All homogenized hazelnut beverage samples exhibited a non-Newtonian flow behavior (n<1) and viscosity values in the flow curves decreased as the shear rate increased, indicating their shear thinning behavior. Additionally, flow behavior of homogenized samples could be well described by the Ostwald de Waele model (R^2>0.977).

The heat treatment intensity caused the significant difference in the viscosity and consistency index of samples at the initial of storage (Table 2). The apparent viscosity of LHT samples was found 0.061 Pa.s while it was found 0.156 Pa.s for HHT samples at the initial of storage. This difference may be due to the protein denaturation and also starch gelatinization during high heat treatment. The presence of aggregates in soy beverage has also been described by Cruz et al. (2007).

Table 2. The rheological changes of LHT and HHT hazelnut beverages during short and long time storage periods, respectively

Sample	Storage (days)	η (Pa.s)	K	n	R^2
LHT	0	0.061±0.005^{a}	0.681±0.02^{a}	0.443±0.028^{b}	0.991
	2	0.044±0.011^{bc}	0.410±0.029^{c}	0.481±0.005^{b}	0.997
	4	0.048±0.008^{b}	0.316±0.016^{d}	0.565±0.008^{a}	0.977
	6	0.046±0.003^{bc}	0.349±0.02^{d}	0.504±0.022^{ab}	0.984
	8	0.038±0.006^{c}	0.393±0.037^{c}	0.488±0.028^{b}	0.985
	10	0.039±0.001^{c}	0.468±0.027^{b}	0.476±0.024^{b}	0.989
HHT	0	0.156±0.013^{c}	7.238±0.18^{c}	0.299±0.017^{b}	0.997
	15	0.257±0.033^{a}	12.707±1.9^{a}	0.345±0.018^{a}	0.995
	30	0.228±0.032^{ab}	9.641±0.75^{b}	0.340±0.029^{a}	0.993
	45	0.235±0.002^{a}	8.73±1.72^{c}	0.330±0.013^{a}	0.994
	60	0.193±0.032^{b}	7.38±0.41^{d}	0.323±0.023^{a}	0.994
	90	0.192±0.021^{b}	8.36±2.69^{c}	0.329±0.036^{a}	0.992
	120	0.207±0.015^{b}	8.37±2.74^{c}	0.335±0.017^{a}	0.989

Values are means ± standard deviation. $^{a-d}$ Means within the same column with different letters are significantly different at $p< 0.05$. η: apparent viscosity at 50 s^{-1} K: consistency index; n: flow behavior index; R^2: determination coefficient of Eq.1

The viscosity and consistency index of low thermal treated hazelnut beverages decreased during the storage. The increase of syneresis value and °Brix could be attributed to the change of rheological behavior during the storage. However, the viscosity and consistency index values of HHT samples increased during the storage. The apparent viscosity was found as 0.207 Pa.s at the end of long time storage. The main reason for the increase of viscosity is the decrease in serum separation values. The denaturation of proteins by heating could have increased the surface hydrophobicity and exposed more sites for hydrophobic interactions with other components, which in turn may have increased the viscosity (Achouri et al., 2007). The flow behavior index of LHT samples did not change during the short time storage however, slight increase was determined for HHT samples after 15 days.

Color properties

Variations in the color values of hazelnut beverages during storage are given in Table 3. Heat treatment intensity did not make any difference for the lightness index of both treated samples on the first day of storage. The L^* value of the LHT samples showed a partial increase up to the 8th day and this increase became more prominent (82.17) in 10 days. However, no change was observed in the a^* and b^* values of the samples during storage. When the total color

difference of LHT hazelnut beverage during storage was examined, it was determined that more color changes occurred at 6th and 10th day of short time storage period, but this change was slightly noticeable to the eye according to the classification described by Cserhalmi et al. (2006). The *L* * value of the HHT sample slightly decreased from 81.33 to 80.71 however, the *a* * value increased from 0.47 to 0.53. The color change of HHT hazelnut beverages compared to the first day became significantly important after 45 days' storage period. At the highest color change observed at the 120th days of storage.

Table 3. The color changes of LHT and HHT hazelnut beverages during short and long time storage periods, respectively

Sample	Storage (days)	*L**	*a**	*b**	ΔE
LHT	0	81.36±0.06^{d}	0.41±0.01^{a}	10.21±0.04^{a}	-
	2	81.66±0.11^{c}	0.32±0.05^{b}	9.92±0.11^{c}	0.37±0.09^{b}
	4	81.75±0.07^{bc}	0.38±0.03^{ab}	10.05±0.08^{bc}	0.39±0.05^{b}
	6	81.68±0.04^{bc}	0.36±0.01^{ab}	10.17±0.06^{a}	0.78±0.04^{a}
	8	81.84±0.07^{b}	0.35±0.03^{ab}	10.19±0.04^{a}	0.43±0.01^{b}
	10	82.17±0.11^{a}	0.36±0.02^{ab}	10.22±0.01^{a}	0.77±0.05^{a}
HHT	0	81.33±0.22^{a}	0.47±0.03^{b}	9.95±0.18^{a}	-
	15	81.08±0.12^{b}	0.45±0.02^{b}	10.1±0.09^{a}	0.29±0.03^{c}
	30	81.25±0.15^{a}	0.47±0.04^{b}	10.04±0.16^{a}	0.35±0.07^{bc}
	45	81.01±0.04^{b}	0.45±0.02^{b}	10.07±0.07^{a}	0.35±0.03^{bc}
	60	81.02±0.09^{b}	0.42±0.01^{b}	9.98±0.02^{a}	0.4±0.02^{b}
	90	80.96±0.05^{b}	0.5±0.02^{a}	10.04±0.08^{a}	0.45±0.03^{b}
	120	80.71±0.15^{c}	0.53±0.03^{a}	10.05±0.03^{a}	0.66±0.04^{a}

Values are means ± Standard Deviation. $^{a-d}$ Means within the same column with different letters are significantly different at $p< 0.05$.

CONCLUSIONS

Physicochemical properties and oxidation stability of hazelnut beverages were affected by both thermal treatment intensity and storage time period. Thermal treatments provoked protein denaturation, thus enhancing the aggregation process. For HHT samples the level of aggregation is higher than LHT samples that caused to decrease in the levels of protein solubility and total soluble matter (Brix). High treatment led to an increase in viscosity and consistency index values of samples compared to low treatment. This could be attributed to increase of the surface hydrophobicity of denatured proteins and exposed more sites for hydrophobic interactions with other components. Syneresis degree was reduced with the increase of heat treatment intensity. Hydroperoxide index showed any difference during the short time storage however, it increased threefold at the end of long time storage period.

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