JOURNAL OF NUTRITION FASTING AND HEALTH

Characterization of White Chocolate Enriched with Free or Encapsulated Pomegranate Extract

Zohreh Didar[1*]
1. Department of Food Science and Technology, Neyshabur Branch, Islamic Azad University, Neyshabur, Iran.

ARTICLEINFO

Article type:
Research Paper

Article History:
Received: 20 Jul 2020
Accepted: 18 Nov 2020
Published: 15 Dec 2020

Keywords:
Pomegranate Extract
White Chocolate
Encapsulation

ABSTRACT

Introduction: Chocolate is a popular food product. White chocolate is produced extensively and has lower functionality compared to other chocolates. The present study aimed to characterize white chocolate enriched with free or encapsulated pomegranate extract.

Methods: To increase the functionality of white chocolate, pomegranate extract was added in the free or encapsulated forms at various concentrations (1, 2, 3, 4, and 5 g/200 g). The coacervation was performed for the encapsulation of pomegranate extract. The quality parameters of the chocolate samples were also evaluated, including the color index, melting behavior, flow behavior (rheometer and scanning electron microscopy), particle size distribution, texture analysis, and organoleptic properties. Other analysis included the determination of the total phenolic compounds and antioxidant properties of the samples.

Results: The addition of pomegranate extract (free/encapsulated) decreased T_{onset}, T_{peak}, and ΔH of white chocolate compared to the control samples. In addition, the added pomegranate extract reduced the casson yield stress, while it increased casson viscosity. The extract also changed the color indices and particle size distribution ($P<0.05$). The white chocolate samples containing pomegranate extract (free/encapsulated) had a higher phenolic content and antioxidant activity than the control samples. Moreover, sensory properties were affected by chocolate formulation, and the samples containing encapsulated pomegranate extract had superior organoleptic properties compared to the samples with free pomegranate extract.

Conclusion: According to the results, free or encapsulated pomegranate extract could enhance the functionality of white chocolate in term of antioxidant activity. Furthermore, organoleptic properties were affected by chocolate formulation, and the samples containing encapsulated pomegranate extract had better organoleptic properties compared to those containing the free form of the extract.

► *Please cite this paper as*:
Didar Z. Characterization of White Chocolate Enriched with Free or Encapsulated Pomegranate Extract. J Nutr Fast Health. 2020; 8(4): 302-309. DOI: 10.22038/jnfh.2020.50603.1281

Introduction

White chocolate is a type of chocolate made of sugar, milk solids, and cocoa butter (1). Due to the absence of cocoa in white chocolate formulation, this chocolate has lower functionality compared to other chocolate types. The enhancement of white chocolate functionality is possible by adding various natural extracts to the formulation, such as the extracts of encapsulated blackberry juice (1), black tea (2), and green tea (3).

Food antioxidants have multiple beneficial effects on human health (4). These components could delay the oxidation rate of foods and increase their freshness and shelf life. Factors such as light, oxygen, temperature, and moisture have debilitative effects on the antioxidant activities of bioactive compounds. Microencapsulation is an approach to the increasing of bioactive stability, as well as the stability of bioactive components. Microencapsulation also modifies the off-flavor, bitter taste, and astringency of polyphenols (5).

Pomegranate (*Punica granatum*) has extensive usage in the folk medicine of many cultures (6). Studies have indicated that pomegranate juice has remarkable antioxidant activity (7), as well as potent antiatherogenic effects on atherosclerotic mice and humans (8). In addition, the abundant phenolic content of pomegranate juice is associated with the prevention of lipid oxidation *in-vitro* (7). Recently, several studies have been focused on the antioxidant, anticarcinogenic, and anti-inflammatory properties of pomegranate constituents, with an emphasis on the treatment and prevention of cancer, cardiovascular diseases, diabetes, dental conditions, erectile dysfunction, bacterial infections, antibiotic resistance, and ultraviolet radiation-induced skin damage. Among the other potential

* *Corresponding author:* Zohreh Didar, Assistant Professor of Food Science, Department of Food Science and Technology, Neyshabur Branch, Islamic Azad University, Neyshabur, Iran. Tel: +58142621908. Fax: +9842615472. E-mail: z_didar57@yahoo.com.
© 2020 mums.ac.ir All rights reserved.
This is an Open Access article distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/3.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

applications of pomegranate are the treatments of male infertility, Alzheimer's disease, arthritis, and obesity (9).

In a study in this regard, Li et al. (2006) analyzed the antioxidant fractions in pomegranate pulp, reporting the mean levels of phenolic compounds (mg/g), flavonoids (mg/g), proanthocyanidins (mg/g), and ascorbic acid (mg/g) to be 24.4±2.7, 17.2±3.3, 5.3±0.7, and 0.85±0.02, respectively (10).

The present study aimed to determine the impact of added pomegranate extract in the free and encapsulated forms on white chocolate formulation and assess the properties of the resulting white chocolate.

Materials and Methods

Experimental Materials

The formulation of the white chocolate samples encompassed sugar, cocoa butter, whole milk powder, sunflower lecithin, and vanilla powder (1). Free and encapsulated pomegranate extracts were added to the samples at the concentrations of 1, 2, 3, 4, and 5 g/200 g. All the chemicals utilized in the present study were purchased from Merck, Germany.

Encapsulation of Pomegranate Extract

The coacervation of pectin and gelatin was performed in suitable conditions for the encapsulation of pomegranate extract. In brief, pectin and gelatin were dissolved in deionized water (60°C, 2 hours), and the biopolymer solutions were centrifuged (5,000 rpm, 30 minutes) for the removal of air bubbles.

At the next stage, a solution comprising of gelatin and pectin (3:1) was prepared (11) and preserved at the temperature of 45°C for 30 minutes and at 300 rpm and proper pH. To complete the formation of microcapsules, an ice-water bath was used for the rapid temperature reduction to below 15°C for 30 minutes at 300 rpm (12). Afterwards, the prepared coacervate capsules were stored at the temperature of 6°C for 24 hours, and the microcapsules were freeze-dried.

Sample Preparation

To prepare white chocolate, 20% of total cocoa butter and dry powders (sugar, whole and skimmed milk powders) were initially blended by gentle heating (40ºC) until the formation of a homogeneous mixture. Afterwards, the chocolate mass was pre-refined on a pilot-scale, three-roll refiner (Lehmann, Aalen, Germany), blended again, and heated to the temperature of 50ºC. Dry conching was carried out at the next stage for 45 minutes. In this step, the rest of the cocoa butter (80% of total) and soy lecithin were also added. Overall, the duration of conching was 360 minutes at the temperature of 60ºC. After conching, free or encapsulated pomegranate extract (1, 2, 3, 4, and 5 g/200 g) was mixed with the chocolate mass at the temperature of 32-33ºC. Following that, the mass was blended for approximately five minutes, and a three-stage tempering process (33-35ºC, 24-25ºC, and 25-26ºC) was performed as well. Molding and vibration processes were also implemented at the temperature of 27-30ºC. The final step was cooling at 5ºC for 20 minutes, and the samples were stored at 13-15ºC in the dark before analysis (13). Table 1 shows the samples and their specific codes.

Table 1. Definition of tested Samples

Samples	Code	Samples	Code
Control (no pomegranate extract)	E0	Control (no pomegranate extract)	E0
White Chocolate Enriched with Free Pomegranate Extract (1 g/200 g)	E1	White Chocolate Enriched with Encapsulated Pomegranate Extract (1 g/200 g)	E6
White Chocolate Enriched with Free Pomegranate Extract (2 g/200 g)	E2	White Chocolate Enriched with Encapsulated Pomegranate Extract (2 g/200 g)	E7
White Chocolate Enriched with Free Pomegranate Extract (3 g/200 g)	E3	White Chocolate Enriched with Encapsulated Pomegranate Extract (3 g/200 g)	E8
White Chocolate Enriched with Free Pomegranate Extract (4 g/200 g)	E4	White Chocolate Enriched with Encapsulated Pomegranate Extract (4 g/200 g)	E9
White Chocolate Enriched with Free Pomegranate Extract (5 g/200 g)	E5	White Chocolate Enriched with Encapsulated Pomegranate Extract (5 g/200 g)	E10

Sample Preparation to Determine the Total Phenolic Content and Antioxidant Activity

To prepare the chocolate samples for determination of the phenolic content, the lipid components were initially removed by adding 10 milliliters of n-hexane to two grams of ground chocolate three times. Residual hexane was also removed by air drying the samples for 24 hours. The phenolic compounds were extracted using an extraction solution composed

of acetone, distilled water, and acetic acid at the ratio of 70:29.8:0.2 v/v. Extraction was performed twice for 30 minutes in an ultrasonic bath (Eurosonic 4D, Italy). After centrifugation (3,000 rpm, one minute), the supernatant phase was decanted and filtered, and the obtained supernatants were mixed to reach 10 milliliters of the extract (14).

Total Phenolic Content

Folin-Ciocalteu reagent was used to determine the total phenolic content of the chocolate samples, and the absorbance was read at 760 nanometers (Jenway 6305 UV/Visible Spectrophotometers, UK). In addition, Gallic acid was used as the standard at various concentrations (62.5-5,000 µmol/l) (15).

Antioxidant Activity

The antioxidant activity of the samples was assessed using the method proposed by Ruiz-Navajas (2013). Accordingly, 100 microliters of the prepared extract solution was blended with four milliliters of DPPH ethanol solution (25 mg/g), and the samples were preserved in the dark for 40 minutes. Following that, the absorbance of the samples was read at the wavelength of 517 nanometers (Jenway 6305 UV/Visible Spectrophotometers, UK). Antioxidant activity was calculated according to the equation (16).

$$I\% = \frac{A_{blank} - A_{sample}}{A_{blank}} \times 100$$

In the equation above, A_{Blank} is the absorption of the control samples, and A_{Sample} shows the absorption of the samples. We used synthetic antioxidant butylated hydroxyanisole (100 ppm) as the standard.

Color Index

The color of the white chocolate samples was determined in terms of L* (lightness), a* (redness to greenness), and b* (yellowness to blueness) using MINOLTA Chroma Meter CR-400 (Minolta Co., Ltd., Osaka, Japan) (1).

Textural Properties of the Chocolate Samples

The textural analysis of the chocolate samples was performed using a texture analyzer (TA. XTPlus, USA) in accordance with the method proposed by Farzanmehr et al. (2008) (18). Before the analysis, the chocolate samples were preserved at the temperature of 20°C for two hours. The texture analyzer also determined the stiffness of the chocolate samples with a flat bottom drill (diameter: 1.6 mm, speed: 90 mm/min, load cell: 500 N). The maximum force at the depth of four millimeters was considered as stiffness (17).

Melting Properties

A differential scanning calorimeter (TA Q20, TA Instruments, New Castle, USA) was used for determining the melting properties of the white chocolate samples (18). The onset temperature (T_{onset}), peak temperature (T_{peak}), end temperature (T_{end}), and energy required for the complete melting of the samples were determined using the resulting thermograms.

Rheological Properties

A stress- or strain-controlled rheometer (MCR 302, Anton Paar, Graz, Austria) was used to evaluate the flow behavior of the chocolate samples at the temperature of 40 ºC. In addition, a cylindrical probe system was applied to determine the rheological parameters based on the following equation:

$$\tau^{0.5} = \tau_0^{0.5} + \eta_{pl.}\,\gamma^{n}$$

where τ is the shear stress (Pa), γ shows the shear rate (s^{-1}), τ_0 is the yield stress (Pa), and η_{pl} represents plastic viscosity (Pa s).

Microstructural Examination

The microstructure of the chocolate samples was observed by scanning electron microscopy (SEM) using a Phenom ProX SEM.

Particle Size Distribution

The particle size distribution of the chocolate samples was assessed using a laser diffraction particle size analyzer (model: Cordouan VASCO 3, France). The determined parameters included d(0.1), d(0.5), and d(0.9), which represented the particle sizes where 10%, 50%, and 90% of the total particle volume was formed by particles smaller than this size, respectively (1).

Sensory Properties

Trained panelists evaluated the impact of the added pomegranate extract on the sensory properties of the chocolate samples, as well as the consumed water and crackers at the interval after each test. Furthermore, acceptance analysis was performed based on the hedonic five-point scale, with five implying complete acceptances, and one implying non-acceptance (19).

Statistical Analysis

All the experiments in the current research were carried out in triplicate. Data analysis was performed in a completely randomized design in

STATISTICA software version 22 using one-way analysis of variance (ANOVA) and Duncan's test at the significance level of 0.05.

Results and Discussion

Thermal Properties and Firmness

Table 2 shows the differential scanning calorimetry (DSC) parameters, including the onset temperature (T_{onset}), peak temperature (T_{peak}), and ΔH. According to the findings, the addition of pomegranate extract (free and encapsulated) influenced the T_{onset} of the chocolate samples, especially at high concentrations, significantly decreasing this parameter compared to the control samples ($P<0.05$). Furthermore, the T_{peak} values of E3, E4, E5, E6, E7, E8, E9, and E10 were significantly lower than the T_{peak} value of the control samples ($P<0.05$). These findings could be attributed to the increased thin layers of the fat phase, which covered a larger specific surface area of the particles in the chocolate samples containing the pomegranate extract. In this regard, Lončarević (2018) reported the reduction of T_{onset} values in the chocolate samples containing encapsulated blackberry juice (1).

Table 2. Melting Properties of White Chocolates Containing Pomegranate Extract (free/encapsulated)

Samples	T_{onset} (ºC)	T_{peak} (ºC)	ΔH (J/g)	Samples	T_{onset} (ºC)	T_{peak} (ºC)	ΔH (J/g)
E0	33±0.1^{a}	35.2±0.1^{a}	3.18±0.2^{a}	E0	32±0.1^{a}	34.5±0.1^{a}	3.13±0.2^{a}
E1	32.6±0.2^{b}	34±0.2^{b}	3±0.5^{b}	E6	31.9±0.2^{a}	34±0.1^{b}	3.1±0.1^{a}
E2	31.1±0.3^{c}	33.7±0.3^{c}	2.6±0.1^{c}	E7	31.5±0.1^{a}	33.6±0.1^{c}	3±0.1^{a}
E3	30.5±0.2^{d}	32.5±0.1^{d}	2.3±0.1^{d}	E8	31.1±0.2^{b}	33.4±0.1^{c}	2.7±0.1^{b}
E4	29.1±0.2^{e}	31.2±0.1^{e}	2.0±.1^{e}	E9	30.2±0.2^{c}	33.0±0.2^{c}	2.5±0.1^{bc}
E5	28.6±0.1^{f}	30±0.2^{f}	1.6±0.2^{f}	E10	29.5±0.1^{d}	32.8±0.1^{d}	2.3±0.1^{c}

**Values expressed as mean±SD of three replicates; superscript lowercase letters show significant differences between samples in column (P<0.05); T_{onset}: onset temperature, T_{peak}: peak temperature, ΔH: required energy for complete melting of samples; experiments performed in triplicate*

According to the results of the DSC analysis, the melting of the control samples occurred at a higher temperature compared to the other samples, which indicated that most of the fat content of the chocolate sample melted above an ambient temperature, thereby leading to the firmer texture of the control samples compared to the other samples. On the other hand, the white chocolate samples containing the pomegranate extract (free/encapsulated) melted at a lower temperature (Table 2).

According to the results of the present study, the T_{peak} values of the chocolate samples containing the pomegranate extract were significantly lower than the controls. However, the required energy for the complete melting of the samples (ΔH) decreased in the samples containing the pomegranate extract as opposed to the control samples; these findings showed the most compact structure in the control samples compared to the enriched samples with the pomegranate extract. This could be attributed to the impact of the extract on fat crystallization and fat network formation in chocolate.

Table 3. Color Indices of White Chocolate Samples

Sample	L*	a*	b*	Sample	L*	a*	b*
E0	71.5±0.02^{a}	−1.18±0.01^{f}	20.22±0.02^{a}	E0	71.5±0.02^{a}	−1.19±0.01^{f}	20.22±0.02^{a}
E1	65.4±0.01^{b}	2±0.001^{e}	18.1±0.03^{b}	E6	67±0.04^{b}	1.5±0.001^{e}	16.1±0.03^{b}
E2	57.1±0.02^{c}	14±0.001^{d}	15±0.01^{c}	E7	62.4±0.04^{c}	11±0.003^{d}	12.2±0.02^{c}
E3	43±0.01^{d}	18±0.001^{c}	12.1±0.02^{d}	E8	58.6±0.04^{d}	14±0.002^{c}	10±0.01^{d}
E4	37.6±0.01^{e}	20±0.002^{b}	9.01±0.02^{e}	E9	46.9±0.02^{e}	16±0.002^{b}	7.01±0.02^{e}
E5	32±0.02^{f}	21±0.002^{a}	6.14±0.02^{f}	E10	41.9±0.02^{f}	18±0.001^{a}	5.1±0.02^{f}

**Values expressed as mean±SD of three replicates; superscript lowercase letters show significant differences between samples in column (P<0.05)*

Color Indices

The color indices in terms of the mean lightness (L*), red tone (a*), and yellow tone (b*) were observed on the surface of the white chocolate samples, and the results are presented in Table 3. Accordingly, the control white chocolate samples had the highest L* (71.5±0.02) and b* values (20.22±0.02), as well as the lowest a* values (-1.18±0.01), compared to the samples containing the pomegranate extract ($P<0.05$), which implied the more bright surface (Table 3). The addition of free pomegranate extract to the white chocolate samples also decreased the L* values (Table 3).

Several factors influence the color indices of chocolate, such as processing parameters,

formulation, and crystal structure (20). In the present study, the addition of the free pomegranate extract to the white chocolate formulation caused more changes in the color indices compared to the samples enriched with the encapsulated pomegranate extract.

Particle Size Distribution

Table 4 shows the parameters associated with the particle size distribution of the white chocolate samples. As can be seen, desirable particles in the chocolate samples were observed within the range of 15-30 micrometers (21). According to our findings, the addition of pomegranate extract to white chocolate formulation changed the particle size of the chocolate samples compared to the control samples ($P<0.05$). Furthermore, the encapsulated pomegranate extract increased the particle size of the chocolate samples.

Table 4. Particle Size Distribution of White Chocolate Samples

Sample	d(0.1)	d(0.5)	d(0.9)	Samples	d(0.1)	d(0.5)	d(0.9)
E0	2.68±0.01^{a}	8.19±0.03^{a}	29.44±0.02^{a}	E0	2.61±0.01^{c}	8.19±0.03^{d}	29.27±0.02^{f}
E1	2.60±0.02^{a}	8±0.03^{a}	28.61±0.04^{b}	E6	2.63±0.02^{c}	8.65±.04^{c}	30.26±0.03^{e}
E2	2.51±0.02^{b}	7.67±0.04^{b}	28.35±.03^{b}	E7	2.67±.03^{c}	9.6±0.04^{b}	32.57±0.02^{d}
E3	2.42±0.01^{c}	7.64±0.04^{b}	27.52±0.04^{c}	E8	2.75±0.03^{b}	9.9±0.03^{b}	35±0.02^{c}
E4	2.35±0.02^{d}	7.64±0.05^{b}	27±0.05^{d}	E9	2.82±0.04^{a}	10.4±0.04^{a}	38.8±0.04^{b}
E5	2.31±0.01^{e}	7.52±0.01^{c}	26.79±0.03^{e}	E10	2.99±0.02^{a}	11.01±0.01^{a}	41.25±0.02^{a}

**Values expressed as mean±SD of three replicates; superscript lowercase letters show significant differences between samples in column ($P<0.05$)*

According to the results of the present study, the addition of the encapsulated pomegranate extracts significantly increased the magnitude of d(0.5) and d(0.9) ($P<0.05$) (Table 4), and the desirable particle size of the chocolate samples was observed within the range of 15-30 micrometers. In general, larger particles than 30 micrometers cause a gritty sensation in the mouth, and particles smaller than 15 micrometers increase the specific surface area, and a higher liquid phase is required for its coverage (21).

Rheological Properties

Table 5 shows the effects of the added pomegranate extract (free/encapsulated) on the rheological properties of the chocolate samples.

Table 5. Rheological Parameters of Enriched White Chocolate

Samples	Casson Yield Stress (Pa)	Casson (Pas)	Samples	Casson Yield Stress (Pa)	Casson (Pas)
E0	6.31±0.1^{a}	0.45±0.02^{c}	E0	6.31±0.1^{a}	0.45±0.1^{b}
E1	6.02±0.11^{b}	0.49±0.01^{b}	E6	6.08±0.13^{b}	0.48±0.12^{a}
E2	5.74±0.12^{c}	0.49±0.01^{b}	E7	5.41±0.11^{e}	0.48±0.13^{a}
E3	5.74±0.1^{c}	0.49±0.01^{b}	E8	5.47±0.12^{d}	0.48±0.11^{a}
E4	5.74±0.12^{c}	0.5±0.01^{a}	E9	5.6±0.12^{cd}	0.48±0.12^{a}
E5	5.42±0.12^{d}	0.51±0.02^{a}	E10	5.69±0.12^{c}	0.48±0.12^{a}

**Values expressed as mean±SD of three replicates; superscript lowercase letters show significant differences between samples in column ($P<0.05$)*

According to the findings of the current research, the addition of pomegranate extract (free/encapsulated) increased the viscosity of the white chocolate mass (Table 5). However, the added encapsulated pomegranate extract had less significant effects on rheological parameters, which is important in terms of technology as the processing of the lower viscosity of chocolate results in easier handling (14). The rheology of chocolate is influenced by various parameters, such as process circumstances, chocolate formulation, fat content, emulsifier, and particle size distribution (22).

Total Phenolic Content and Antioxidant Activity of the Chocolate Samples

Table 6 shows the measurement of the total phenolic content of the chocolate samples with or without added pomegranate extract. Accordingly, the addition of pomegranate extract (free/encapsulated) increased the phenolic content of the chocolate samples (Table 6). As the content of the added pomegranate extract increased, the content of phenolic compounds increased as well.

Previous findings have indicated that pomegranate juice has high antioxidant activity (7) and several beneficial health effects (8), which are associated with the presence of

various phenolic compounds in pomegranate juice. These compounds are able to scavenge free radicals and prevent lipid oxidation *in-vitro* (6). In the current research, the analysis of the antioxidant activity indicated the enhanced antioxidant activity in the white chocolate samples enriched with pomegranate extract (free/encapsulated). This observation is in accordance with the increased phenolic content of the chocolate samples following the addition of the pomegranate extract.

Table 6. Total Phenolic Content (mg/100 g) and Antioxidant Activity of White Chocolate Samples

Sample Code	Total Phenolic Content (mg/100 g)	Antioxidant Activity (%)
E0	3.6±0.5^{i}	5.1±2.1^{i}
E1	17.4±1.8^{h}	16.2±2.2^{h}
E2	26.4±0.9^{e}	19.1±4.1^{f}
E3	28.2±2.2^{d}	21.69±2.8^{d}
E4	29.2±1.3^{c}	23.2±4.1^{c}
E5	31.1±1.2^{a}	25.2±2^{a}
E0	3.6±0.5^{h}	5.1±2.1^{i}
E6	18.02±10.5^{g}	18.2±3.2^{g}
E7	25.1±1.4^{f}	20.1±3.2^{e}
E8	29.25±2.5^{c}	23.1±4.1^{c}
E9	30.18±3.4^{b}	24.3±2.5^{b}
E10	31.15±1.4^{a}	25.1±1.2^{a}

**Values expressed as mean±SD of three replicates; means with different letters show significance in column (P≤0.05)*

Textural Properties of the Chocolate Samples

The textural properties of the chocolate samples were analyzed, and the results were reported in terms of stiffness (18). Table 7 shows the results of the texture analysis of the white chocolate samples in term of stiffness.

Table 7. Stiffness of White Chocolate Samples

Sample Code	Stiffness
E0	59114.1±550^{a}
E1	49524.8±456^{d}
E2	48427.5±326^{e}
E3	43464.3±222^{f}
E4	42681.5±211^{g}
E0	59114.1±550^{a}
E6	53259.3±429^{b}
E7	51656.9±242^{c}
E8	49682.4±452^{d}
E9	43355.2±784^{f}
E10	42236.2±684^{g}

**Values expressed as mean±SD of three replicates; means with different letters show significance in column (P≤0.05)*

Table 8. Organoleptic Properties of White Chocolate Samples

Sensory Properties	E0	E1	E2	E3	E4	E5	E6	E7	E8	E9	E10
Texture	3.9±0.00^{a}	3.5±0.03^{b}	3±0.02^{c}	2.8±0.02^{d}	2.5±0.04^{f}	2.3±0.03^{g}	3.8±0.02^{a}	3.5±0.03^{b}	3±0.01^{c}	2.9±0.02^{c}	2.6±0.02^{e}
Taste	3.7±0.02^{ab}	3±0.02^{c}	2.8±0.01^{d}	2.5±0.01^{e}	2±0.01^{f}	1.9±0.01^{f}	3.9±0.02^{a}	3.7±0.02^{ab}	3.5±0.03^{b}	3±0.01^{c}	2.8±0.00^{d}
Melting in Mouth	3.8±0.02^{a}	3±0.01^{c}	3.5±0.04^{b}	3.2±0.01^{bc}	3±0.03^{c}	2.8±0.03^{d}	3.5±0.04^{b}	3±0.02^{c}	3±0.03^{c}	3±0.01^{c}	2.9±0.01^{d}
Apparent Color	3.9±0.01^{a}	3.5±0.00^{b}	3±0.01^{c}	2.8±0.02^{d}	2.5±0.02^{e}	2±0.02^{f}	3.7±0.01^{ab}	3.2±0.03^{c}	3.5±0.01^{b}	2.8±0.01^{d}	2±0.01^{f}
Overall Acceptance	3.9±0.01^{a}	3.6±0.03^{b}	3±0.02^{c}	3.5±0.02^{b}	3±0.03^{c}	3.8±0.01^{a}	3.7±0.03^{a}	3.8±0.02^{a}	3±0.01^{b}	3.5±0.02^{b}	3±0.02^{c}

**Superscript lowercase letters show significant differences between samples (P<0.05)*

According to the information in Table 7, the addition of pomegranate extract affected the texture of the resulting chocolate samples, and a significant difference was observed between the enriched samples with pomegranate extract (free/encapsulated) and the control samples in this regard (P<0.05). In both cases, the stiffness of the chocolate samples was lower than the controls (Table 7).

Appearance and Microstructural Properties

In the present study, the SEM technique was performed to assess the microstructure of chocolate (Figure 1). With the exception of the sample containing 5 g/200 g of the free

pomegranate extract, the other chocolate samples had a uniform and homogenous structure.

Organoleptic Properties of the Chocolate Samples

According to our findings, the addition of pomegranate extract (free/encapsulated) had a significant impact on the chocolate color; the samples containing the free pomegranate extract had a darker color, while the samples incorporated with the encapsulated pomegranate extract had less color change. In both cases, color intensity increased at the higher concentrations of the added pomegranate extract. Notably, the highest score of color belonged to the control samples. In addition, the chocolate surface glow was affected by the added pomegranate extract, and both sample groups had a lower score than the control samples in this regard.

A B

Figure 1. SEM Images of White Chocolate; A) Free Pomegranate Extract (5 g/200 g), B) Encapsulated Pomegranate Extract (5 g/200 g)

The panelists stated that the addition of pomegranate extract reduced the required time for chocolate melting in the mouth. The addition of the encapsulated pomegranate extract increased the hardness of chocolate, while the free pomegranate extract resulted in lower hardness compared to the control samples. Furthermore, the addition of the free or encapsulated pomegranate extract had a significant impact on the chocolate flavor ($P<0.05$), while the effect of the encapsulated pomegranate extract was less significant on the taste of chocolate compared to the free pomegranate extract.

Conclusion

According to the results, free and encapsulated pomegranate extract could enhance the functionality of white chocolate in term of antioxidant activity. Moreover, the addition of both forms of the pomegranate extract decreased the T_{onset}, T_{peak}, and ΔH values of white chocolate. While casson yield stress decreased, casson viscosity increased with the addition of pomegranate extract. The particle size and color indices were also influenced by the added pomegranate extract ($P<0.05$). Finally, organoleptic properties were affected by the chocolate formulation, and the samples containing the encapsulated pomegranate extract showed better sensory properties compared to the samples containing the free pomegranate extract.

Conflicts of interest

None declared.

Acknowledgments

Hereby, we extend our gratitude to the director of the central laboratory of the Islamic Azad University of Neyshabur Branch for assisting us in this research project.

References

1. Lončarević I, Pajin B, Fišteš A, Tumbas Šaponjac V, Petrović J, Jovanović P, Vulić J, Zarić D. Enrichment of white chocolate with blackberry juice encapsulate: Impact on physical properties, sensory characteristics and polyphenol content. LWT - Food Sci. Technol 2018; 92:458–64.
2. Lončarević I, Pajin B, Petrović J, Danica Zarić D, Šaponjac VT, Fišteš A, Petar Jovanović P. The physical properties, polyphenol content and sensory characteristics of white chocolate enriched with black tea extract. Food in Health and Disease. 2019; 8(2):83-8.
3. Lončarević I, Pajin B, Šaponjac VT, Petrović J, Vulić J, Fišteš A, Jovanović P. Physical, sensorial and bioactive characteristics of white chocolate with encapsulated green tea extract. Quality and shelf life of white chocolate with green tea encapsulate. J Sci Food Agric. 2019; 99(13): 5834-41.
4. Ozkan G, Franco P, De Marco I, Xiao J, Capanoglu E. A review of microencapsulation methods for food

antioxidants: Principles, advantages, drawbacks and applications. Food Chem. 2019; 272:494–506.
5. Ballesteros L F, Ramirez M J, Orrego C E, Teixeira J A, Mussatto S I. Encapsulation of antioxidant phenolic compounds extracted from spent coffee grounds by freeze-drying and spray-drying using different coating materials. Food Chem. 2017; 237:623-31.
6. Longtin R. The pomegranate: nature's power fruit?. J Natl *Cancer* Inst. 2003; 95(5): 346–8.
7. Gil M I, Tomas-Barberan F A, Hess-Pierce B, Holcroft D M, Kader A A. Antioxidant activity of pomegranate juice and its relationship with phenolic composition and processing. J Agric Food Chem. 2000; 48(10):4581–9.
8. Aviram M, Dornfeld L, Rosenblat M, Volkova N, Kaplan M, Coleman R, et al. Pomegranate juice consumption reduces oxidative stress, atherogenic modifications to LDL, and platelet aggregation: studies in humans and in atherosclerotic apolipoprotein E-deficient mice. Am J Clin Nutr 2000; 71(5):1062–76.
9. Jurenka J. Therapeutic applications of pomegranate (*Punica granatum L.):* a review. Altern Med Rev. 2008; 13(2):128-44.
10. Li Y, Guo C, Yang J, Wei J, Xu J, Shuang Cheng S. Evaluation of antioxidant properties of pomegranate peel extract in comparison with pomegranate pulp extract. Food Chem 2006; 96(2):254–60.
11. Duhoranimana E, Karangwa E, Lai Lingfeng, Xu, Xing, Yu, Jingyang, Xia, Shuqin, et al. Effect of sodium carboxymethyl cellulose on complex coacervates formation with gelatin: Coacervates characterization stabilization and formation mechanism. Food Hydrocolloids. 2017; 69:111–20.
12. Muhoza B, Xia S, Cai J, Zhang X, Duhoranimana E, Su J. Gelatin and pectin complex coacervates as carriers for cinnamaldehyde: Effect of pectin esterification degree on coacervate formation, and enhanced thermal stability. Food Hydrocolloids. 2019; 87: 712–22.
13. Toker OS, Konar N, Rasouli Pirouzian H, Oba S, Genc Polat D, Palabiyik I, Poyrazoglu, ES, Sagdic O. Developing functional white chocolate by incorporating different forms of EPA and DHA - Effects on product quality. LWT - Food Sci. Technol. 2018; 87: 177-85.
14. Todorovic V, Redovnikovic IR, Todorovic Z, Jankovic G, Dodevska M, Sobajic S. Polyphenols, methylxanthines, and antioxidant capacity of chocolates produced in Serbia. J Food Compos Anal. 2015; 41:137-43.
15. Singleton VL, Orthofer R, Lamuela-Ravent, RM. Analysis of total phenols and other oxidation substrates and antioxidants by means of folin-ciocalteu reagent. Methods Enzymol 1999; 299: 152-78.
16. Ruiz-Navajas Y, Viuda-Martos M, Sendra E, Perez-Alvarez J A, Fernández-López J. In vitro antibacterial and antioxidant properties of chitosan edible films incorporated with *Thymus moroderi* or *Thymus piperella* essential oils. J Food Control. 2013; 30(2):386–92.
17. Farzanmehr H, Abbasi S, Sahari MA. Effect of sugar replacer on some physicochemical, rheological and sensory properties of milk chocolate. Iran J Nutr Sci Food Technol. 2008; 3(3):65–82. [in Persian]
18. Glicerina V, Balestra F, Rosa M D, Romani S. Rheological, textural and calorimetric modifications of dark chocolate during process. J. Food Eng. 2013; 119(1): 173-79.
19. Botelho PB, Galasso M, Dias V, Mandrioli M, Lobato LP, Rodriguez-Estrada MT. Castro IA. Oxidative stability of functional phytosterol-enriched dark chocolate. LWT- Food Sci Technol. 2014; 55(2): 444-51.
20. Lindecrantz A. Investigation of seedpowder technology for pre-crystallization processing for dark chocolate- Effect on fat crystal structure and storage stability. 2014; Master of Science Thesis in the Master Degree Program Biotechnology. Gothenburg, Sweden: Chalmers University of Technology.
21. Bolenz S, Holm M, Langkrär C. Improving particle size distribution and flow properties of milk chocolate produced by ball mill and blending. Eur. Food Res. Technol 2014; 238(1):139–47.
22. Pajin B, Dokić Lj, Zarić D, Šoronja-Simović D, Lončarević I, Nikolić I. Crystallization and rheological properties of soya milk chocolate produced in a ball mill. J. Food Eng. 2013; 114(1):70–4.