



Using paprika extract in chocolate spread and white compound chocolate: effects on color stability and bioavailability

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Abstract

White compound chocolate (CC) and chocolate spread (CS) do not contain cocoa solids and thus they have deficiency of antioxidant activity. So, paprika extract (PE) was used to develop the visual and functional features of compound chocolate (CC) and chocolate spread (CS). Color stability of both chocolate samples was investigated for 12 weeks under accelerated shelf-life conditions and using PE significantly increased a^* , C^* , and h° values of chocolate samples ($P < 0.05$). Also, at the end of shelf life, ΔE values were determined as > 2.83 and > 3.74 for CC and CS samples, respectively. Using PE in the production of CC and CS samples significantly increased antioxidant activity for pre- and post in vitro digestion, except for post digestion of S025 samples ($P < 0.05$). General acceptability of the CS samples decreased with increasing of PE ratios, for CC samples it was increased except for C100. The results showed that PE can be used in the production of functional chocolate with high antioxidant activity and as a natural coloring agent.

Keywords Chocolate · Paprika · Color stability · Bioavailability · Sensory evaluation

Introduction

Carotenoids are antioxidant compounds that are in charge of the color of numerous fruits and vegetables and which have beneficial effects on human welfare [1]. Paprika is a spice produced from the peels of *Capsicum annuum* and it is rich in carotenoids, and about half of these carotenoids are capsanthine, but also contains capsorubin, violaxanthin, zeaxanthin, β -cryptoxanthin and β -carotene [2]. Paprika extract, the earliest and the most considerable inherent carotenoid food colorants, is used in the production of foods and beverages

[3]. One of the main approaches in functional food development studies is to add bioactive compounds to foods [4]. So, paprika may be an important potential in the development of functional food and as a food additive due to the antioxidant and positive effects on health of its carotenoid compounds [5]. Bioavailability of bioactive components of foods could be linked on several factors like food formulation, food matrices, kind of nutrients, process conditions and as well as gastrointestinal conditions [6]. Carotenoids have lipophilic characteristics and chocolate is formed from fat emulsions [6], so chocolate formulation could affect bioavailability of carotenoids. Therefore, it will be considerable to identify the bioavailability of these compounds used in functional food development studies.

The confectionery industry, especially in relation to cocoa products like chocolate, has gained active alterations within last decades, and the needing for innovative food produced with natural ingredients is increasing day by day [7]. Therefore, it is essential to identify the main characteristics of functional chocolate products developed with natural ingredients, such as color, water activity (a_w), moisture content, texture, flow behavior, melting profile, rheology and sensory analysis [8]. On the other hand, a limited number of works on white chocolate containing natural ingredients such as paprika extract have been carried out. Mainly studies on the

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white chocolate concern: rheological characteristics [9, 10], color loss kinetics [11], flavonoid content [12], their acute effect on coronary circulation [13], browning [14], increase in flow features [15], and the interaction of white chocolate casein and phenolic compounds during the storage process [16]. This study, hence, will fill the gap about functional white chocolate containing natural ingredients in the confectionery literature and industry as well.

This study aimed to develop functional white compound chocolates and chocolate spreads containing natural paprika extract. So, effect of using paprika extract on the principal characteristics of chocolate products such as a_w values, moisture content, color, texture, flow behavior, melting profile, rheology and sensory analysis was investigated to determine the acceptability of developed products by consumers. Furthermore, total antioxidant capacities of paprika extract and functional chocolate samples produced with using paprika extract were determined under in vitro conditions for before and after digestion and bioavailabilities were calculated.

Materials and methods

Chocolate spread and compound chocolate production

Using paprika extract chocolate spread and compound chocolate samples were manufactured properly to the our previous study [17]. Chocolate samples were produced by considering the same ingredients, production steps and ratios.

a_w values and moisture content

One day after the production, the a_w value of each chocolate samples (CS and CC) was determined at room temperature (25 °C) by using Lab-Master a_w (Novasina, Switzerland) [18]. The moisture content value of chocolate products (CS and CC) and components were obtained with the methodology explained by [19].

Color properties

Color characteristics (L^* , a^* and b^*) of chocolate products (CS and CC) were determined with colorimeter device (Chroma Meter CR-400, Konica Minolta, Japan). Equations 1, 2, 3 and 4 were used to calculate the chroma (C^*), hue (h°), whiteness index (WI^*) and color stability (ΔE) of the samples, respectively [20]. The CS and CC products were stored after the production under accelerated shelf-life (ASL) situations (25 °C–75% RH) [20]. The color parameters of the products were measured at weekly intervals for 12 weeks.

$$C^* = \sqrt{a^{*2} + b^{*2}} \quad (1)$$

$$h^\circ = \arctan(b^*/a^*) \quad (2)$$

$$WI = 100 - [(100 - L)^2 + a^2 + b^2]^{1/2} \quad (3)$$

$$\Delta E = \left[(DL^*)^2 + (Da^*)^2 + (Db^*)^2 \right]^{1/2} \quad (4)$$

Sensory evaluation

The sensorial assessment of CS and CC samples were realized in accordance with the procedure defined before by [17] with 40 panelists (25 females, 15 males, 18–33 age). The goal of the work was clarified to the panelists with an information text. A confirmation form was signed by the panelists, if they accepted the aim of the study. Before assessment, each panelists was trained about sensorial analysis, chocolate samples and methodology. Asked to the panelists for cleaning their mouth with water between each sample. The panelists were asked to scored the samples from 1 to 5 by using the Multiple Comparison Technique, taking into account the sensorial parameters represented in the Fig. 3. All CS and CC samples were scored in 4 meetings consecutively in 5 days.

Texture analysis

To determine the hardness value of the CC samples, a texture analyzer was used and methodology was adapted previously described by [18]. Analysis was done 5 replicates and results were stated as the average value. Also, a method previously mentioned by [21] was used to perform the texture features of the CS samples.

Melting behavior

The melting parameters of the CC products were specified with DSC (TA Q20, USA) in accordance with the procedure informed by [22]. T_{onset} , T_{peak} , T_{end} and Δh (J/g) values of the products were figured out from thermograms.

Rheological properties

The rheological analysis of the CC products was applied with using a temperature controlled rheometer (Anton Paar, MCR 302, Austria) and the procedure defined by [23] was applied. Using the below expressed equation (Eq. 5) the Casson model parameters were calculated:

$$\tau^{0.5} = \tau_0^{0.5} + \eta_{pl} \dot{\gamma}^n \quad (5)$$

τ , $\dot{\gamma}$, τ_0 and η_{pl} have indicated shear stress (Pa), shear rate (s^{-1}), yield stress (Pa) and plastic viscosity (Pa s), respectively.

Sample extraction

Methanol was used as a solvent for extraction of the CC, CS and PE samples and extraction was carried out with ultrasonication and Ultraturrax (IKA Labortechnik, Staufen, Germany) in accordance with the methodology declared by [24].

Antioxidant activity

DPPH radical scavenger method was used to determination of antioxidant capacity of the samples [25]. 0.1 mL of the extract was diluted in certain ratios and was mixed with prepared 2.9 mL of DPPH radical solution and 6×10^{-5} mM methanol. The solution was stored in the darkness place for 25 min at room temperature and the absorbance values were obtained at 517 nm. Gallic acid was used as a standard and using created calibration curve DPPH antioxidant capacity of the products was calculated.

Determination of bioavailability

The total antioxidant capacity of pre-digested and in vitro post-digested CC and CS samples and PE were analyzed for Bioavailability investigation. The Bioavailability of these two parameters was tested as well. The pre-digestion antioxidant determination of the CC and CS samples was conducted by means of a chemical extraction method illustrated in 2.9. Digestion simulation was determined with a method described by [26]. Briefly, 5 g of ground CC or CS samples were taken into centrifugal tube and mixed with 5 mL of the oral fluid. About 12 mL of purified water was added and were hold in a water bath for at 37 °C for 4–5 min. After the completion of the mouth digestion 10 mL of stomach fluid was put into the mix and incubated in the shaking incubator for 2 h at 37 °C. pH of the samples was set to neutral pH values by addition of bile salts. This mixture was mixed with 10 mL of intestine fluid and hold in water bath for 2 h at 37 °C. After pancreatic digestion, 50 mL distilled water added to the samples and centrifuged (13,000 rpm, 5 min). The resulting fluid was carefully taken out and filtered by a 0.45 μ m filter. The DPPH capacity of the samples was calculated with gallic acid standard. The bioavailability of the samples was given as the ratio of the result found by the chemical extraction method to the result obtained by the in vitro digestion simulation method.

Statistical analysis

Statistical differences of the samples were determined using MINITAB 16.0 software programme. The data were given as average value and standard deviation. ANOVA was performed to specify statistical importance and significance level was determined with Tukey's test ($P < 0.05$).

Results and discussion

Water activity and moisture content

The moisture content and a_w values of chocolate products are important parameters due to the shelf life and impact on hardness, viscosity and melting properties, and also chocolate products has low moisture content and water activity [27]. As shown in Fig. 1, a_w and moisture content (g/100 g) of CC and CS samples were found as 0.25–0.37 and 0.31–0.45, and 0.77–2.10 and 1.46–1.80 (g/100 g), respectively. Using PE at various levels resulted high water activity and moisture content for both CC and CS samples, but only moisture content of CS samples found to be statistically unimportant. The a_w values of the samples were in agreement with the values stated in the literature (0.3 and 0.5) [19, 27]. Also, the moisture content of the samples were in agreement with literature except C100 sample. The increase in a_w and moisture content of chocolate samples is related to the use of paprika extract. However, there

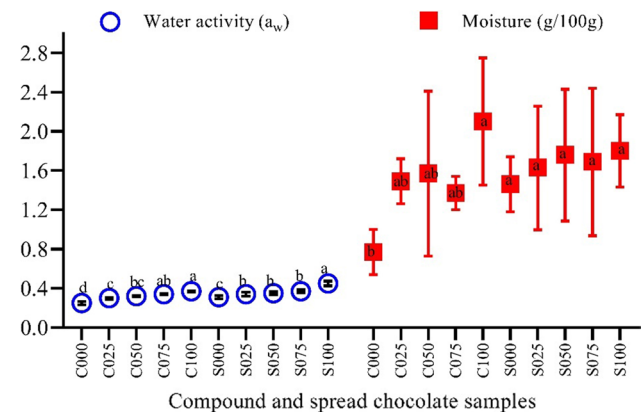


Fig. 1 Water activity and moisture content of compound and spread chocolate samples. Different superscript lowercase letters show the significant differences for each sample group ($P < 0.05$). All experiments were performed in triplicate. All data are represented as means \pm standard deviation. (C000 and S000: Compound and Spread Chocolate Control, C025 and S025: Compound and spread chocolate including 0.25 paprika extract (g/100 g), C050 and S050: Compound and Spread Chocolate including 0.50 paprika extract (g/100 g), C075 and S075: Compound and Spread Chocolate including 0.75 paprika extract (g/100 g), C100 and S100: Compound and Spread Chocolate including 1.0 paprika extract (g/100 g))

was no negative increase in the a_w and moisture content of all samples at a level that would have affected the quality parameters of the samples. [17] reported that using beetroot powder and black carrot extract didn't significantly affect the a_w and moisture content of chocolate samples, whereas [28] reported that using different plant extract significantly affected the moisture content of dark chocolate.

Color properties and stability

Color not only affects sensory characteristics, but also affects perceptions and attitudes in chocolate samples [29, 30]; hence, it is a parameter that should be taken into account when producing new chocolate samples [30]. Using PE in the production of CC and CS samples resulted lower L^* , h° and WI values, while a^* , b^* and C^* values of the samples increased (Fig. 2). Generally, chocolate products have middle or extended shelf life [31]. However, white chocolate and its products contain less antioxidant activity compared to other chocolate categories, they may tend to lose color during shelf life [32, 33]. Therefore, substances containing natural antioxidant compounds such as paprika may contribute to the shelf life of white chocolate. All chocolate samples were stored for 12 weeks under ASL conditions and color properties were determined weekly, with each week under ASL conditions representing a period of 1 month [20]. Since the ΔE value above 3.0 represents a noticeable color change in the samples [34], the ΔE values of all samples were calculated (Table 1). ΔE values of CC and CS samples were determined as between 0.6–3.16 and 0.86–4.96, respectively. However, ΔE values above 3.0 were not found, except for a few samples of CC

and CS. As a result, using PE in the production of CC and CS samples ensured favorable color stability throughout samples' shelf life.

Sensory features

Appearance, flavour, aroma, texture, color are important parameters for consumer acceptance [35]. In this work, the sensorial parameters, shown in the Fig. 3, were evaluated by the panelists for all chocolate samples. In the CC samples, using PE resulted better appearance and color properties set against to the control and it was found that the general acceptability scores of all CC samples were similar compared to the control. However, although a very small difference was determined in terms of melting in the mouth and buying intense, no significant difference was determined in terms of brightness, sweetness, odor-taste and aroma. Using PE at high ratio (1 g/100 g) in the production of CS samples resulted negatively in terms of appearance, melting in mouth, taste, buying intense and general acceptance. However, using PE at other ratios (0.25, 0.50 and 0.75 g/100 g) in the production CS samples resulted better scores. This study has shown that although the use of PE at different ratios may cause negative effects on some sensory properties of CS samples, these can be ignored or improved, and thus PE can be used in the production of CC and CS samples with high sensory liking. In parallel to our study, [10] reported that the use of blackberry extract in white chocolate production did not affect negatively sensory features of chocolate samples.

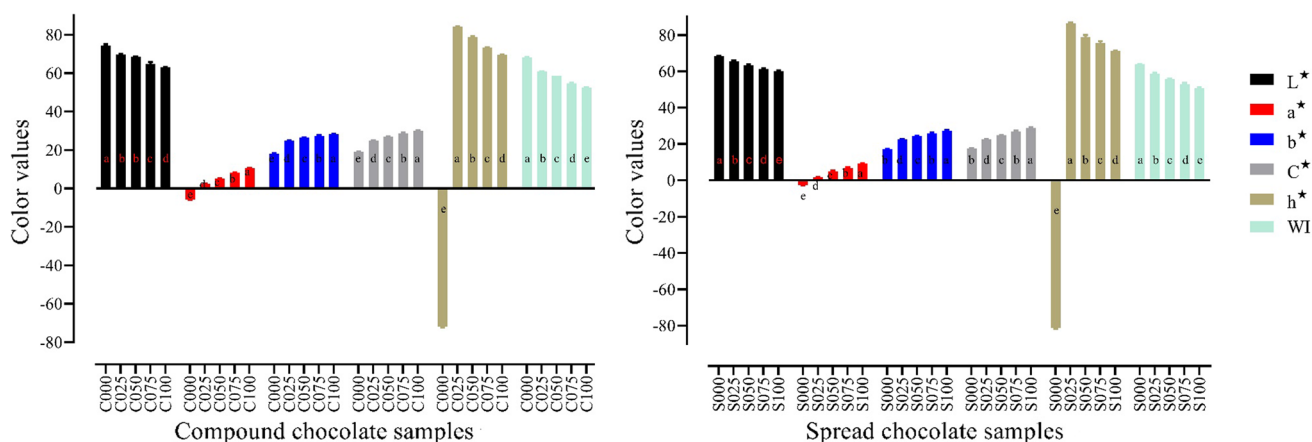


Fig. 2 Color properties of compound and spread chocolate samples after production. WI: Whiteness index. Different superscript lowercase letters show the significant differences for each sample group ($P < 0.05$). All experiments were performed in triplicate. All data are represented as means \pm standard deviation. (C000 and S000: Compound and Spread Chocolate Control, C025 and S025: Compound

and Spread Chocolate including 0.25 paprika extract (g/100 g), C050 and S050: Compound and Spread Chocolate including 0.50 paprika extract (g/100 g), C075 and S075: Compound and Spread Chocolate including 0.75 paprika extract (g/100 g), C100 and S100: Compound and Spread Chocolate including 1.0 paprika extract (g/100 g))

Table 1 Color stability of compound chocolate samples under accelerated shelf life conditions

Sample	Storage time (weeks)												
	0	1	2	3	4	5	6	7	8	9	10	11	12
C000													
L*	74.4±0.65 ^{bc}	75.5±0.48 ^a	75.5±0.51 ^b	74.0±0.54 ^{ab}	74.4±0.64 ^{ab}	74.6±0.89 ^{ab}	74.4±0.59 ^{ab}	73.7±0.89 ^{ab}	74.3±0.54 ^{ab}	74.0±0.88 ^{ab}	73.2±0.02 ^b	73.2±0.93 ^b	73.6±0.66 ^{ab}
a*	-5.9±0.06 ^g	-5.2±0.05 ^f	-4.9±0.03 ^e	-4.8±0.06 ^e	-4.7±0.09 ^{de}	-4.5±0.15 ^{cd}	-4.4±0.08 ^c	-4.3±0.11 ^{bc}	-4.2±0.04 ^{ab}	-4.1±0.12 ^{ab}	-4.1±0.06 ^a	-4.0±0.11 ^a	-4.0±0.06 ^a
b*	18.2±0.17 ^g	18.4±0.11 ^{fg}	18.2±0.28 ^g	18.4±0.32 ^{efg}	18.9±0.15 ^{defg}	18.3±0.29 ^{fg}	19.2±0.17 ^{cd}	19.5±0.23 ^{bc}	18.8±0.24 ^{def}	19.1±0.32 ^{cde}	20.3±0.13 ^a	20.3±0.37 ^a	20.9±0.21 ^{ab}
ΔE	nd	1.34±0.40 ^{fg}	1.42±0.33 ^{efg}	1.29±0.15 ^g	1.49±0.13 ^{efg}	1.67±0.14 ^{defg}	1.85±0.03 ^{cdef}	2.30±0.09 ^{bc}	1.89±0.01 ^{cde}	2.18±0.10 ^{cd}	2.99±0.13 ^a	3.16±0.14 ^a	2.83±0.09 ^{ab}
C025													
L*	69.7±0.34 ^{cd}	70.8±0.45 ^a	70.7±0.31 ^{ab}	70.5±0.11 ^{abc}	70.3±0.09 ^{abc}	70.8±0.43 ^a	70.2±0.19 ^{abc}	70.4±0.34 ^{abc}	70.4±0.43 ^{abc}	70.2±0.12 ^{abc}	68.6±0.53 ^e	69.7±0.29 ^{bcd}	68.9±0.11 ^{de}
a*	2.54±0.11 ^d	2.57±0.11 ^d	2.54±0.16 ^d	2.59±0.12 ^d	2.64±0.11 ^{cd}	2.86±0.14 ^{abcd}	3.10±0.26 ^b	3.09±0.16 ^{ab}	2.70±0.20 ^{bcd}	2.88±0.06 ^{abcd}	3.26±0.18 ^a	2.80±0.13 ^{bcd}	3.05±0.07 ^{abc}
b*	24.8±0.14 ^e	25.5±0.38 ^b	25.9±0.16 ^{ab}	25.9±0.13 ^{ab}	25.9±0.16 ^{ab}	25.8±0.17 ^{ab}	25.7±0.30 ^{ab}	26.2±0.13 ^a	24.2±0.10 ^c	24.5±0.08 ^e	25.7±0.38 ^{ab}	26.0±0.25 ^{ab}	25.9±0.15 ^{ab}
ΔE	nd	1.33±0.58 ^{ab}	1.55±0.13 ^a	1.43±0.03 ^{ab}	1.30±0.18 ^{ab}	1.60±0.38 ^a	1.25±0.25 ^{ab}	1.69±0.21 ^a	0.97±0.22 ^{abc}	0.69±0.09 ^e	1.69±0.24 ^a	1.30±0.27 ^{ab}	1.45±0.10 ^a
C050													
L*	68.5±0.15 ^{bc}	69.2±0.16 ^a	68.3±0.41 ^{abcd}	68.3±0.19 ^{bcd}	68.3±0.14 ^{bcd}	68.8±0.22 ^{ab}	68.4±0.47 ^{abcd}	68.4±0.13 ^{bcd}	68.7±0.27 ^{abc}	68.2±0.53 ^{bcd}	67.1±0.22 ^e	67.8±0.38 ^{cde}	67.6±0.43 ^{de}
a*	5.22±0.14 ^a	5.02±0.28 ^a	4.84±0.29 ^a	4.91±0.23 ^a	4.97±0.17 ^a	5.22±0.21 ^a	5.23±0.58 ^a	5.57±0.22 ^a	4.93±0.20 ^a	5.30±0.06 ^a	5.62±0.22 ^a	4.95±0.35 ^a	5.09±0.18 ^a
b*	26.4±0.14 ^{ab}	27.3±0.34 ^a	27.0±0.34 ^a	27.1±0.22 ^a	27.1±0.11 ^a	26.9±0.23 ^a	27.0±0.75 ^a	27.4±0.09 ^a	25.3±0.13 ^c	25.8±0.38 ^{bc}	27.0±0.24 ^a	27.2±0.47 ^a	26.4±0.48 ^{ab}
ΔE	nd	1.20±0.28 ^{ab}	1.67±0.24 ^a	1.22±0.21 ^{ab}	1.06±0.45 ^{ab}	0.87±0.05 ^{abc}	0.84±0.03 ^{abc}	0.81±0.08 ^{abc}	0.60±0.26 ^c	0.96±0.50 ^{abc}	1.13±0.15 ^{ab}	1.18±0.16 ^{ab}	0.78±0.58 ^c
C075													
L*	64.9±0.86 ^{bcd}	66.6±0.16 ^a	65.8±0.54 ^{abcd}	66.0±0.25 ^{abc}	66.2±0.12 ^{ab}	66.2±0.39 ^{ab}	66.4±0.26 ^a	66.2±0.15 ^{abc}	66.3±0.75 ^a	66.3±0.13 ^a	64.5±0.36 ^d	65.9±0.09 ^{abc}	64.9±0.77 ^{cd}
a*	8.22±0.08 ^{bcde}	8.18±0.18 ^{bcd}	8.01±0.13 ^{cde}	8.06±0.10 ^{cde}	8.12±0.08 ^{cde}	8.40±0.09 ^{abcd}	8.66±0.40 ^{ab}	8.53±0.03 ^{abc}	7.75±0.25 ^e	7.98±0.26 ^{de}	8.74±0.10 ^a	7.94±0.12 ^{de}	7.93±0.22 ^{de}
b*	27.4±0.49 ^b	28.8±0.22 ^a	28.4±0.39 ^a	28.7±0.23 ^a	29.0±0.08 ^a	28.6±0.16 ^a	28.2±0.16 ^{ab}	28.9±0.10 ^a	26.5±0.55 ^c	26.5±0.45 ^c	28.4±0.19 ^{ab}	28.2±0.13 ^{ab}	28.4±0.35 ^a
ΔE	nd	2.16±0.26 ^a	1.29±0.58 ^{bcd}	1.67±0.30 ^{abc}	2.05±0.11 ^{ab}	1.72±0.40 ^{abc}	1.81±0.26 ^{abc}	1.90±0.17 ^{abc}	1.92±0.23 ^{abc}	1.73±0.23 ^{abc}	1.20±0.11 ^{bcd}	1.29±0.07 ^{bcd}	1.18±0.18 ^c
C100													
L*	63.1±0.15 ^{def}	65.0±0.61 ^{ab}	64.4±0.20 ^{abc}	64.4±0.11 ^{abc}	64.3±0.33 ^{abc}	62.8±0.11 ^{ef}	62.4±0.47 ^f	64.1±0.40 ^{bc}	65.1±0.27 ^a	64.2±0.39 ^{abc}	62.5±0.19 ^f	64.0±0.22 ^{cd}	63.5±0.38 ^{cde}
a*	10.5±0.12 ^{ab}	10.4±0.18 ^{abc}	10.5±0.06 ^{abc}	10.4±0.05 ^{abc}	10.3±0.13 ^{abc}	9.2±0.12 ^d	10.5±0.27 ^{ab}	10.7±0.17 ^a	9.8±0.44 ^{cd}	9.8±0.30 ^{bcd}	10.7±0.24 ^a	10.2±0.27 ^{abc}	10.1±0.40 ^{abc}
b*	28.2±0.15 ^{bc}	29.6±0.22 ^a	29.7±0.09 ^a	29.7±0.14 ^a	29.7±0.31 ^a	26.0±0.24 ^d	28.6±0.18 ^{ab}	29.5±0.31 ^a	27.1±0.34 ^{cd}	26.8±0.31 ^d	28.5±0.13 ^{ab}	28.8±0.36 ^{ab}	28.5±0.42 ^{ab}
ΔE	nd	2.36±0.58 ^{ab}	2.02±0.19 ^{abc}	1.94±0.16 ^{abc}	1.87±0.43 ^{abc}	2.60±0.16 ^a	1.35±0.28 ^{bcd}	1.63±0.49 ^{abcd}	2.44±0.45 ^a	1.91±0.30 ^{abc}	0.76±0.05 ^e	1.12±0.25 ^{cde}	0.74±0.56 ^{de}
S000													
L*	68.4±0.16 ^a	68.6±0.94 ^a	67.4±0.22 ^a	67.9±0.75 ^a	67.0±1.63 ^a	67.4±1.98 ^a	67.8±1.06 ^a	66.8±1.64 ^a	68.1±1.57 ^a	67.1±0.97 ^a	66.7±0.67 ^a	66.8±1.07 ^a	66.2±1.37 ^a
a*	-2.61±0.11 ^f	-2.44±0.22 ^g	-2.06±0.05 ^f	-1.76±0.02 ^{ef}	-1.46±0.02 ^{de}	-1.24±0.11 ^{cd}	-1.04±0.05 ^{abc}	-1.21±0.26 ^{bcd}	-1.05±0.09 ^{abc}	-1.03±0.10 ^{abc}	-0.85±0.08 ^a	-0.92±0.07 ^{ab}	-1.09±0.04 ^{abc}
b*	17.3±0.06 ^d	17.7±0.06 ^{cd}	17.9±0.58 ^{de}	18.5±0.49 ^{bcd}	19.1±0.55 ^{ab}	18.1±0.45 ^{cde}	19.2±0.22 ^{ab}	19.1±0.53 ^a	18.1±0.25 ^{cde}	18.4±0.14 ^{cde}	19.7±0.28 ^a	19.6±0.40 ^{ab}	18.2±0.29 ^{bcd}
ΔE	nd	0.92±0.20 ^{ef}	1.36±0.20 ^{de}	1.72±0.37	2.89±0.35 ^{ab}	2.45±0.58 ^{abcd}	2.67±0.08 ^{abc}	3.11±0.51 ^{ab}	2.21±0.27 ^{bcd}	2.44±0.33 ^{abcd}	3.47±0.32 ^a	3.38±0.19 ^a	2.93±0.85 ^{ab}
S025													
L*	65.6±0.42 ^{ab}	66.9±0.35 ^a	64.6±1.06 ^{abc}	64.6±0.56 ^{abc}	64.6±0.21 ^{abc}	65.2±0.17 ^{ab}	64.3±0.85 ^{abc}	64.9±0.82 ^{abc}	64.3±1.53 ^{abc}	61.9±2.72 ^c	63.8±0.52 ^{bc}	64.0±0.75 ^{abc}	64.0±0.15 ^{abc}
a*	1.46±0.19 ^h	1.80±0.20 ^{gh}	2.00±0.24 ^{gh}	2.28±0.08 ^{efg}	2.55±0.22 ^{def}	3.01±0.18 ^{bcd}	3.05±0.27 ^{bcd}	3.54±0.24 ^{ab}	2.87±0.31 ^{cde}	3.44±0.25 ^{abc}	3.73±0.28 ^a	3.19±0.15 ^{abcd}	3.32±0.08 ^{abc}
b*	22.5±0.25 ^{cd}	23.3±0.46 ^{bcd}	23.7±0.40 ^{abc}	24.0±0.12 ^{ab}	24.2±0.15 ^{ab}	24.4±0.21 ^{ab}	24.7±0.38 ^a	24.9±0.44 ^a	22.1±1.22 ^d	23.3±0.12 ^{bcd}	24.8±0.32 ^a	24.3±0.31 ^{ab}	24.4±0.21 ^{ab}
ΔE	nd	1.33±0.06 ^{cd}	1.89±0.25 ^{bcd}	2.07±0.17 ^{bcd}	2.26±0.23 ^{bcd}	2.48±0.21 ^{bc}	3.06±0.26 ^{abc}	3.34±0.40 ^{ab}	2.35±1.06 ^{bc}	4.55±1.99 ^a	3.70±0.12 ^{ab}	3.05±0.17 ^{abc}	3.08±0.22 ^{abc}
S050													
L*	63.5±0.30 ^b	64.5±0.35 ^a	62.1±0.45 ^c	62.0±0.31 ^c	62.0±0.06 ^c	61.5±0.35 ^{cd}	60.8±0.32 ^{de}	60.8±0.56 ^{de}	60.9±0.42 ^{de}	60.4±0.35 ^e	58.9±0.17 ^f	60.2±0.06 ^e	60.1±0.15 ^e
a*	4.79±0.51 ^{efg}	4.47±0.21 ^g	4.34±0.03 ^g	4.46±0.04 ^g	4.56±0.08 ^{fg}	5.05±0.19 ^{def}	5.14±0.05 ^{cde}	5.82±0.05 ^{ab}	5.69±0.24 ^{abc}	5.65±0.09 ^{abc}	6.15±0.01 ^a	5.61±0.11 ^{abc}	5.56±0.17 ^{bcd}
b*	24.4±0.06 ^f	25.1±0.21 ^{de}	25.1±0.38 ^e	25.2±0.17 ^{cde}	25.3±0.10 ^{cde}	25.6±0.06 ^{bcd}	25.7±0.10 ^{bcd}	26.4±0.06 ^a	25.7±0.46 ^{bcd}	25.2±0.10 ^{abc}	25.7±0.00 ^{bcd}	25.9±0.06 ^{ab}	25.7±0.15 ^{bc}
ΔE	nd	1.27±0.31 ^f	1.66±0.28 ^{de}	1.72±0.19 ^{de}	1.77±0.11 ^{de}	2.28±0.28 ^{cd}	3.00±0.27 ^{bc}	3.51±0.40 ^b	3.01±0.50 ^{bc}	3.24±0.30 ^b	4.96±0.17 ^a	3.76±0.03 ^b	3.74±0.18 ^b

Table 1 (continued)

Sample	Storage time (weeks)													
	0	1	2	3	4	5	6	7	8	9	10	11	12	
S075														
L*	61.4±0.35 ^b	63.4±0.36 ^a	60.7±0.30 ^{ab}	60.3±0.23 ^{bcd}	60.3±0.12 ^{bcd}	60.3±0.15 ^{bcd}	60.7±0.45 ^{bc}	59.4±0.21 ^{def}	59.3±0.32 ^{def}	60.0±0.15 ^{ede}	59.6±0.20 ^{cdef}	58.7±1.11 ^f	59.2±0.20 ^{def}	59.1±0.20 ^{ef}
a*	6.70±0.64 ^{bc}	7.11±0.02 ^{abc}	6.75±0.15 ^d	6.75±0.09 ^{bc}	6.75±0.25 ^{bc}	6.74±0.44 ^{bc}	6.96±0.40 ^{abc}	7.26±0.33 ^{abc}	7.88±0.22 ^a	7.45±0.11 ^{ab}	7.40±0.18 ^{ab}	6.33±0.25 ^c	7.74±0.33 ^a	7.26±0.27 ^{ab}
b*	25.9±0.47 ^d	26.3±0.45 ^{cd}	26.7±0.15 ^{bcd}	26.7±0.15 ^{bcd}	26.9±0.12 ^{abc}	27.2±0.15 ^{abc}	26.6±0.36 ^{bcd}	27.3±0.25 ^{ab}	27.7±0.27 ^a	26.9±0.21 ^{abc}	26.4±0.20 ^{cd}	26.6±0.25 ^{bcd}	27.1±0.21 ^{abc}	26.6±0.38 ^{bcd}
ΔE	nd	2.45±0.22 ^{ab}	1.37±0.26 ^{bcd}	1.52±0.18 ^{bcd}	1.68±0.11 ^{bcd}	1.68±0.11 ^{bcd}	1.07±0.43 ^{cd}	2.46±0.34 ^{ab}	2.98±0.47 ^a	1.93±0.25 ^{abc}	2.01±0.25 ^{abc}	2.10±0.30 ^{abc}	3.01±1.12 ^a	2.58±0.33 ^{ab}
S100														
L*	60.0±0.36 ^{bc}	60.7±0.30 ^{ab}	59.1±0.36 ^{bcd}	58.9±0.10 ^{ede}	58.9±0.10 ^{ede}	58.7±0.12 ^{ede}	58.7±0.27 ^{ede}	59.4±0.95 ^{bcd}	58.2±0.06 ^{de}	61.7±0.51 ^a	57.9±0.31 ^{de}	57.3±1.36 ^e	58.2±0.53 ^{de}	58.0±0.15 ^{de}
a*	9.27±0.10 ^{cd}	9.2±0.15 ^d	9.29±0.06 ^{cd}	9.28±0.05 ^{cd}	9.28±0.05 ^{cd}	9.27±0.05 ^{cd}	9.49±0.09 ^{bcd}	9.43±0.30 ^{cd}	9.88±0.10 ^{ab}	9.88±0.15 ^{ab}	9.65±0.09 ^{abc}	10.0±0.18 ^a	9.62±0.21 ^{abc}	9.92±0.16 ^a
b*	27.4±0.25 ^{de}	28.3±0.06 ^a	28.1±0.06 ^{ab}	28.1±0.06 ^{ab}	28.1±0.06 ^{ab}	28.1±0.06 ^{ab}	27.9±0.21 ^{abc}	27.6±0.21 ^{cd}	28.2±0.10 ^{ab}	26.3±0.12 ^f	27.1±0.06 ^e	27.3±0.21 ^{de}	27.9±0.06 ^{bc}	28.1±0.15 ^{ab}
ΔE	nd	1.20±0.17 ^{bcd}	1.25±0.29 ^{bcd}	1.37±0.08 ^{bcd}	1.49±0.14 ^{abcd}	1.49±0.14 ^{abcd}	1.43±0.30 ^{bcd}	0.86±0.82 ^d	2.12±0.11 ^{abc}	2.07±0.44 ^{abc}	2.31±0.23 ^{ab}	2.84±1.15 ^a	1.92±0.48 ^{abc}	2.20±0.11 ^{6abc}

nd not determined

Different superscript lowercase letters show the significant differences for each sample group due to storage time ($P < 0.05$). All experiments were performed in triplicate. All data are represented as means ± Standard deviation. (C000: Compound chocolate control, C025: Compound chocolate including 0.25 paprika extract (g/100 g), C050: Compound chocolate including 0.50 paprika extract (g/100 g), C075: Compound chocolate including 0.75 paprika extract (g/100 g), C100: Compound chocolate including 1.0 paprika extract (g/100 g))

Textural properties

Hardness, which is one of the principal textural features of chocolate products, can be linked with the ingredients of the chocolate and the production steps [36] and it should be important in chocolate manufacturing [37]. As seen in the Fig. 4, the hardness values of CC samples were found between as 4413–8141 g and using PE in the production CC of samples resulted higher hardness values and the results were found to be statistically important compared to the control ($P \leq 0.05$). [10] stated that blackberry extract can have an important effect on the hardness values of white chocolate and that increasing the extract concentration increases the hardness values. The textural features of the chocolate spread, shown in Fig. 4, are closely related to the sensory properties [38]. It was determined that these parameters changed between 588 and 8868 g, (-611)-(-1669) g/min, (-268)-(-476) and 480–7504 g/min g, respectively and using PE in the production of chocolate spread increased textural values of CS samples ($P \leq 0.05$). It is found that although using of PE at low ratios did not cause a significant change, using PE at high ratios increased these quality parameters and this may be related to the using PE in liquid form.

Flow behaviour

Rheological features of chocolate are effective on getting high quality products and inform about sensorial properties of chocolate [39]. The yield stress and plastic viscosity values of the CC samples were calculated with obtained data of shear stress and shear rate by adapting to the Casson model, and R^2 values were indicated the compatibility of the obtained data with the Casson model (Table 2) and these values of control sample and CC samples including PE were determined in the range of 1.68–6.93 Pa and 2.09–3.81 Pa s, respectively. It was determined that using PE in the production of CC samples importantly increased yield stress and plastic viscosity of the samples ($P \leq 0.05$). These results are in accordance with moisture content, as the increasing moisture increases rheological parameters [39]. [10] reported similar results to our study, and it was stated that the use of blackberry extract in white chocolate production significantly raised the yield stress values of the chocolate samples ($P \leq 0.05$).

Melting behaviour

Melting behavior of chocolate is a considerable parameter that affects consumer behavior [22]. Melting parameters of T_{onset} , T_{peak} , T_{end} and Δh were determined as 20.7–22.0 °C, 31.9–33.1 °C, 40.2–40.8 °C, and 33.2–35.3 j/g, respectively by using DSC technique and statistically significant weren't found ($P > 0.05$) (Table 2). However, only the

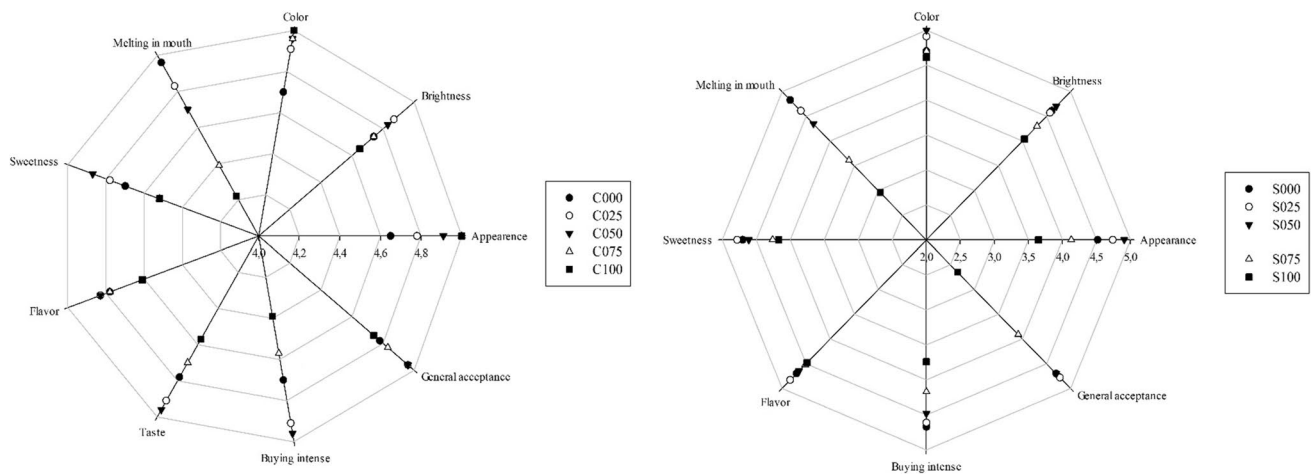


Fig. 3 Sensorial evaluation of compound chocolate and spread chocolate samples. (C000 and S000: Compound and Spread Chocolate Control, C025 and S025: Compound and Spread Chocolate including 0.25 paprika extract (g/100 g), C050 and S050: Compound and

Spread Chocolate including 0.50 paprika extract (g/100 g), C075 and S075: Compound and Spread Chocolate including 0.75 paprika extract (g/100 g), C100 and S100: Compound and Spread Chocolate including 1.0 paprika extract (g/100 g))

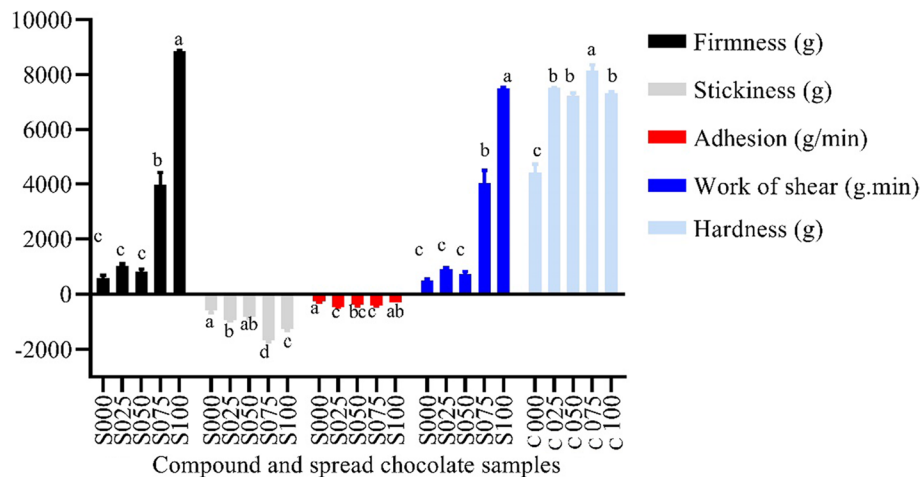


Fig. 4 Texture properties of compound and spread chocolate samples. Different superscript lowercase letters show the significant differences for each sample group ($P < 0.05$). All experiments were performed in triplicate. All data are represented as means \pm standard deviation. (C000 and S000: Compound and Spread Chocolate Control, C025 and S025: Compound and Spread Chocolate includ-

ing 0.25 paprika extract (g/100 g), C050 and S050: Compound and Spread Chocolate including 0.50 paprika extract (g/100 g), C075 and S075: Compound and Spread Chocolate including 0.75 paprika extract (g/100 g), C100 and S100: Compound and Spread Chocolate including 1.0 paprika extract (g/100 g))

T_{onset} value of C050 and C100 products were determined as statistically important ($P \leq 0.05$). Contrary to our study [10] reported that using blackberry extract in white chocolate production resulted in significantly lower T_{onset} values compared to the control sample ($P \leq 0.05$). This situation may be related to the using different formulations of chocolate samples (which we developed compound chocolate and spreadable chocolate), the different extracts used, or the different methodology used to determine the melting profile.

Bioavailability properties

One of the main goals of this study is to increase antioxidant activity of white CC and CS samples by using PE as a natural coloring agent. Since, white chocolate derivatives are lack of antioxidant activity because it does not contain cocoa mass and solids. In this study, CC and CS samples were subjected to in vitro digestion of the mouth, stomach and intestine and antioxidant activities were determined by using DPPH method and values were

Table 2 Flow and melting behaviour and pre- and post-digestion antioxidant activity of compound and spread samples including paprika extract

Samples	Antioxidant activity			Flow behaviour			Melting behaviour			
	Before digestion	After digestion	Bioavailability (%)	σ_0 (Pa)	η_{pl} (Pa.s)	R^2	T_{onset} (°C)	T_{peak} (°C)	T_{end} (°C)	Δh (J/g)
Paprika extract	16.3 ± 4.301	18.9 ± 2.260	115.6							
C000	0.078 ± 0.006 ^b	0.124 ± 0.009 ^d	159.1	1.68 ± 0.25 ^c	2.09 ± 0.01 ^d	0.995	21.3 ± 0.38 ^{ab}	32.0 ± 0.26 ^a	40.8 ± 0.83 ^a	35.3 ± 1.55 ^a
C025	0.111 ± 0.009 ^c	0.140 ± 0.001 ^d	125.9	2.22 ± 0.13 ^c	2.81 ± 0.09 ^c	0.990	21.4 ± 0.26 ^{ab}	32.2 ± 0.50 ^a	40.4 ± 0.38 ^a	34.2 ± 0.57 ^a
C050	0.133 ± 0.003 ^c	0.208 ± 0.002 ^c	157.1	2.22 ± 0.16 ^c	3.36 ± 0.11 ^b	0.986	20.7 ± 0.54 ^b	33.1 ± 1.58 ^a	40.4 ± 0.93 ^a	33.5 ± 4.40 ^a
C075	0.218 ± 0.004 ^b	0.286 ± 0.001 ^b	131.2	3.95 ± 0.24 ^b	3.73 ± 0.03 ^a	0.987	21.8 ± 0.15 ^{ab}	31.9 ± 0.23 ^a	40.3 ± 1.03 ^a	33.2 ± 3.78 ^a
C100	0.249 ± 0.002 ^a	0.360 ± 0.006 ^a	144.9	6.93 ± 1.14 ^a	3.81 ± 0.26 ^a	0.963	22.0 ± 0.04 ^a	32.0 ± 0.00 ^a	40.2 ± 0.11 ^a	35.2 ± 1.47 ^a
S000	0.105 ± 0.007 ^c	0.269 ± 0.015 ^c	255.8	–	–	–	–	–	–	–
S025	0.141 ± 0.007 ^d	0.229 ± 0.0055 ^d	162.5	–	–	–	–	–	–	–
S050	0.151 ± 0.001 ^c	0.312 ± 0.0055 ^c	206.3	–	–	–	–	–	–	–
S075	0.222 ± 0.002 ^b	0.486 ± 0.0022 ^b	218.7	–	–	–	–	–	–	–
S100	0.257 ± 0.002 ^a	0.552 ± 0.002 ^a	215.0	–	–	–	–	–	–	–

Different superscript lowercase letters show the significant differences for each sample group ($P < 0.05$). All experiments were performed in triplicate. All data are represented as means ± standard deviation. Total antioxidant activity values of before and after digestion were expressed as mg Trolox Equivalent/g. (C000 and S000: Compound and Spread Chocolate Control, C025 and S025: Compound and Spread Chocolate including 0.25 paprika extract (g/100 g), C050 and S050: Compound and Spread Chocolate including 0.50 paprika extract (g/100 g), C075 and S075: Compound and Spread Chocolate including 0.75 paprika extract (g/100 g), C100 and S100: Compound and Spread Chocolate including 1.0 paprika extract (g/100 g))

given as gallic acid equivalent (mg Trolox Equivalent/mL) (Table 2). Also, bioavailabilities were calculated by comparing the obtained results. As expected, antioxidant activities of CC and CS samples increased with increasing PE concentration, and the highest antioxidant value was determined for all samples at the highest concentration of PE extract (1 g/100 g). In general, the antioxidant activity of the samples were increased more than 2 times at used the highest PE concentration compared to control sample. In addition, it was found that the antioxidant values of all samples in terms of gallic acid equivalent increased for post-digestion compared to pre-digestion. The antioxidant activity of both CC and CS samples was found to be statistically significant in parallel with the increase in PE concentration ($P \leq 0.05$). Although it has been reported that the utilization of plant extract in chocolate production has improved the antioxidant activity of chocolate samples [10, 17, 40–42], some studies have reported that antioxidant activity may decrease due to the use of plant extract [43, 44]. This may be depend on the extraction method, the chocolate formulation or the type of plant extract used.

Conclusion

Carotenoids are significant food ingredients due to their both coloring and bioactivity effects. The effect of colorants used in functional product development on the quality and visual properties of foods are important. In this study, the use of PE as a source of carotenoids in CC and CS samples was investigated. Results showed that using PE extract there was no significant effect on the physicochemical, texture, flow, and melting features of CC and CS products. The use of PE was dispersed homogeneously in fat-phase chocolate without the need for encapsulation and metal chelation techniques. CC and CS samples have shown sufficient color stability throughout their shelf life. In addition, the use of PE extract in the production of CC and CS samples increased the antioxidant activities of the samples. Finally, sensory evaluation showed that PE extract can be used in the production of CC and CS samples with high consumer appeal.

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Author contributions HG: software, writing—original draft. AB: investigation, formal analyses. NK: project administration. MY: data curation. OS: project administration.

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Data availability The data that support the findings of this study are available from the corresponding author upon reasonable request.

Declarations

Conflict of interest The authors declare no conflict of interest.

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