

Original article

Bittersweet chocolates containing prebiotic and sweetened with stevia (*Stevia rebaudiana* Bertoni) with different Rebaudioside A contents: multiple time–intensity analysis and physicochemical characteristicsBruna Marcacini Azevedo,^{1*} Janaína Madruga Morais-Ferreira,¹ Valdecir Luccas² & Helena Maria André Bolini¹

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Summary The objective of this study was to evaluate the time–intensity profile of the sensory attributes possibly affected in sugar-free and low-fat chocolates containing inulin and stevia with different rebaudioside A contents, such as sweetness, bitterness and melting rate. The bittersweet chocolates were analysed by the multiple time–intensity analysis. The time–intensity profile for the sweetness stimulus was similar for all chocolate samples. The differences between the contents of rebaudioside A were not perceived by the assessors. In relation to the bitterness stimulus, the low-fat samples had a more accentuated perception of this attribute by the assessors, with significant differences for I_{max} and Area when compared to the sugar-free samples. The stimulus melting in the mouth was more affected in the low-fat samples. The sensory results obtained in this study are useful for food industry and researchers working with sweeteners and prebiotics in food, especially in chocolates.

Keywords Chocolate, glycosides, multiple time–intensity analysis, prebiotics, sensory analysis.

Introduction

In recent years, there is a growing demand for natural products and foods with nutritionally balanced profile and healthy ingredients. Health and physical fitness, along with cultural and ethical concerns, has led to new food choices (Falguera *et al.*, 2012). Therefore, researchers have been focused on reformulating food products traditionally rich in saturated fat and sugar, by the replacement with natural ingredients with positive physiological effects, including natural sweeteners and prebiotic compounds. Several studies have changed the traditional bar chocolate formulations into healthier alternatives while maintaining the sensory characteristics (Azevedo *et al.*, 2016; Morais-Ferreira *et al.*, 2016; Saputro *et al.*, 2017).

Stevia rebaudiana is a sweetener that has gained much attention, being the only natural non-nutritive

sweetener approved in Europe (Philippe *et al.*, 2014). In addition, *Stevia rebaudiana* Bertoni leaves have high concentrations of steviol glycosides, including stevioside and rebaudioside A. Despite its commercial use, the stevioside has residual bitter taste, which limits its applications in the food industry. The number of carbohydrate units at positions C-13 and C-19 determines the degree of the component's sweetness. Rebaudioside A has an extra unit of glucose at position C-13 when compared to stevioside, presenting greater properties in terms of sweetness and flavour profile, thus becoming a potential component in stevia (Zhang & Li, 2013). The higher commercial demand has led to the need to increase the rebaudioside A content of stevia leaves, which has been the target of many studies (Ramesh *et al.*, 2006; Rajasekaran *et al.*, 2007; Yadav *et al.*, 2014; Zhang *et al.*, 2015).

Sugar replacement for a high-intensity sweetener, such as stevia, can cause serious changes in chocolate manufacturing. Therefore, a technological alternative to solve this problem may be the addition of fibres,

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such as inulin. The incorporation of inulin into food products presents numerous technological advantages, including texturising, thickening, gelling and emulsifying properties, besides being sugar and fat substitutes (Shourideh *et al.*, 2012). In addition, the consumption of inulin is associated with health benefits, such as inhibition of colon cancer, immunomodulatory effects, reduction in the risk of cardiovascular diseases (Barclay *et al.*, 2010), relief in constipation (Shoaib *et al.*, 2016) and increase in mineral absorption (Al-Sheraji *et al.*, 2013), among others.

Sensory perception during food consumption is a dynamic phenomenon (Cliff & Heymann, 1993). The time-intensity (TI) analysis is a methodology widely used to obtain the intensity variations of a specific attribute over time and has been successfully applied to several matrices, including chocolate (Morais-Ferreira *et al.*, 2016), French fries (Luckett *et al.*, 2016), beverages (Sokolowsky *et al.*, 2015), chocolate milk containing chia oil (Rodrigues *et al.*, 2015) and meat products (Lorido *et al.*, 2014). Multiple time-intensity analysis (MTIA) is a way to graphically represent simultaneously the dynamic profiles of two or more sensory attributes of a single sample (Morais *et al.*, 2014).

The main objective of this study was to evaluate the time-intensity profile of the sensory attributes possibly affected in sugar-free and low-fat chocolates containing inulin and stevia with different rebaudioside A contents.

Materials and methods

Material

The following ingredients were used to prepare the bittersweet chocolate formulations: stevia extract with different rebaudioside A (Reb A) contents (60%, 80% and 97%) (Steviafarma Industrial, Maringá, Brazil); Orafit GR[®] inulin (Beneo, Mannheim, Germany); maltitol (Sweet Pearl[®] P90 Roquette, Roquette Freres, France); icing sugar (Mais Doce, Limeira, Brazil); cocoa mass (Barry Callebaut, Extrema, Brazil); deodorised cocoa butter (Barry Callebaut, Extrema, Brazil); skimmed milk powder (Embaré, Lagoa da Prata, Brazil); Solec CH soy lecithin (Solae, Esteio, Brazil); Grinsted[®] – PGPR 90 polyglycerol polyricinoleate (Danisco, Cotia, Brazil); and artificial vanilla flavour (Synergy Aromas, Vinhedo, Brazil).

Methods

Bittersweet chocolate manufacturing

All samples were produced in the Cereal and Chocolate Technology Center (CEREAL CHOCOTEC) of

the Food Technology Institute (ITAL) located in Campinas, SP, Brazil.

The chocolates were conventionally produced. The ingredients were homogenised in a Kitchen-Aid model K5SS planetary mixer (Kitchen-Aid, St. Joseph, MI, USA) and subsequently refined in a Draiswerk GMBH three-roll refiner (Draiswerke GmbH, Mannheim Waldo, Mahweh, NJ, USA). After the conching step (65 °C/16 h) in an Inco longitudinal conch (Inco, Avaré, SP, Brazil), the samples were tempered on a marble table. The chocolates were placed in polycarbonate moulds with bar shape, and subjected to vibration on a Jafinox (BR) vibratory table. The moulds were then passed twice through a cooling tunnel (Siaht, Jundiá, SP, Brazil). The samples were wrapped in aluminium foil and stored for 15 days at 20 °C in a BOD incubator (Novatecnica, BR, Brazil).

Seven formulations were prepared (Table 1), as follows: a conventional chocolate (containing sugar), 3 stevia-sweetened chocolates (using stevia with 60%, 80% and 97% rebaudioside A) and 3 stevia-sweetened chocolates with a reduction of 50% cocoa butter. A previous study was carried out to determine the equivalent sweetness of each sweetener as compared with sucrose (Azevedo *et al.*, 2016).

According to the Brazilian legislation (Brasil, 2016), the amount of inulin in the chocolate samples of this study was sufficient to guarantee the prebiotic activity of this fibre.

Sensory evaluation

Chocolate samples (4 g) were presented to the tasters in monadic form with three replicates in disposable plastic cups encoded with 3-digit numbers (Meilgaard *et al.*, 1999), using a complete balanced block design (Stone *et al.*, 2012). Crackers and taste-free water were provided for palate cleansing.

The sensory tests were performed in individually air-conditioned booths (22 °C) in the Sensory Science and Consumer Studies Laboratory of the Food Engineering Faculty (FEA) at University of Campinas (UNICAMP), Brazil. All computers were equipped with the software Time-Intensity Analysis of Food and Tastes (TIAFT) (Unicamp, 2012).

Time-intensity analysis

The time-intensity analysis was performed for the attributes sweetness, bitterness and melting rate, which can be directly affected in sugar-free, low-fat chocolates.

The individuals were recruited among undergraduate and graduate students and staff from the University of Campinas, bittersweet chocolate lovers interested in

Table 1 Formulation of bittersweet chocolates

Ingredients (%)	Sucrose	Chocolate samples					
		Stevia 60%	Stevia 80%	Stevia 97%	Stevia 60%	Stevia 80%	Stevia 97%
		Reb A	Reb A	Reb A	Reb A*	Reb A*	Reb A*
Icing sugar	47.5	0	0	0	0	0	0
Cocoa mass	30	30	30	30	30	30	30
Cocoa butter	15	15	15	15	7.5	7.5	7.5
Maltitol	0	32.34	32.34	32.34	32.34	32.34	32.34
Inulin	0	15	15	15	20	20	20
Skimmed milk powder	6.9	6.9	6.9	6.9	9.4	9.4	9.4
Soy lecithin	0.3	0.3	0.3	0.3	0.3	0.3	0.3
PGPR	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Vanilla flavour	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Stevia	0	0.16	0.16	0.16	0.16	0.16	0.16

*Low-fat samples.

participating in the study. Only individuals with the ability to work with computers were selected at this stage.

Triangle tests were applied in the preselection of the tasters, using the Wald sequential analysis (Meilgaard *et al.*, 1999). Two commercial samples of bittersweet chocolate with different cocoa contents, with a significant difference of 0.1% previously determined in a paired test, were used. The variable values used in Wald's sequential analysis were as follows: $r_0 = 0.45$, $r_1 = 0.70$, $a = 0.05$ and $b = 0.05$. Seventeen individuals were preselected in this step.

Training and selection of the time-intensity analysis team

The seventeen preselected individuals were trained for the attributes sweetness, bitterness and melting rate, and the references are shown in Table 2. The training aimed to form the sensory memory on the maximum perception for each stimulus and consisted in the direct contact of the individuals with the reference of

Table 2 Definition and references of the stimuli for bittersweet chocolates evaluated by the assessors

Attributes	Definition	Reference
Sweetness	Taste stimulated by the presence of sucrose and other compounds, such as sweetener	Bittersweet chocolate containing 49% sucrose (produced in CEREAL CHOCOTEC of ITAL)
Bitterness	Characteristic taste of caffeine	Bittersweet chocolate 43% (Lacta Amaro [®])
Melting rate	Time required for solid chocolate turning into liquid while moving the tongue	Aired milk chocolate (Nestlé Suflair Duo [®])

maximum intensity occurring for each stimulus, during three sessions of 1h each.

The conditions for the time-intensity analysis were standardised as follows: (i) judge's wait time of 10 s; (ii) time with the sample in the mouth of 15 s; (iii) time after swallowing of 90 s; and (iv) nine intensity scales. The scale used for performing the tests was linear and horizontal from 0 to 9, in which 0 corresponded to none (far left), 4.5 corresponded to moderate (middle) and 9 corresponded to strong (far right) (Palazzo & Bolini, 2014).

To perform the analysis, after hearing the signal given by the computer (10 s), the tasters were instructed to place the entire sample (4 g) in the mouth and indicate the intensity of perception according to time, by sliding the mouse over the scale.

At the end of the test, the following parameters were collected and analysed statistically: maximum intensity (I_{max} , the maximum sweetness intensity of each sample), time for maximum intensity ($T_{I_{max}}$, the maximum intensity time), area under the curve (Area, total area under the time-intensity curve) and total time (T_{tot} , time for the perception of sweetness from the first to the last perception).

For the selection of the final team, each taster evaluated the stimulus separately, and twelve assessors of the seventeen preselected individuals were chosen to participate according to their discriminative ability ($P < 0.30$), repeatability ($P > 0.05$) and group consensus (Damasio & Costell, 1991).

Multiple time-intensity analysis

Twelve selected assessors (seven females and five males) evaluated all attributes (sweetness, bitterness and melting in the mouth) of the monadic samples, according to a complete block design (Wakeling &

MacFie, 1995), with three repetitions. The attributes were evaluated separately.

Physicochemical characterisation

Texture profile

The texture profile of all chocolate samples was performed at the Central Laboratory of the Department of Food and Nutrition (Unicamp/FEA). Samples were evaluated in five replicates. A Texture Analyzer TA-XT2, Stable Micro Systems, with the software Texture Expert for Windows, was used, which measured the compression force of all samples. The probe comprised two metallic, parallel bars and a third bar coupled to the machine arm, which descended vertically in predetermined speeds, causing sample fracture. The test conditions were as follows: pretest speed: 3.0 mm s⁻¹; test speed: 1.7 mm s⁻¹; post-test speed: 10.0 mm s⁻¹; rupture test distance: 5.0 mm; distance: 20.0 mm; force: 0.981 N; and time: 5.0 s.

The mean hardness values were expressed as compressive force (g), and the standard deviations were calculated.

Colour measurements

The colour of the bittersweet chocolates was determined according to the CIELab system. The parameters L* (brightness), a* (redness) and b* (yellowness) were measured in a spectrophotometer (Color Quest II model, Hunter Associates Laboratory, Reston, VA, USA), adjusted with the following parameters: 10° angle, illuminant D65 and calibration mode RSIN (Minolta, 1994).

Statistical analysis

The parameters collected by the time-intensity curves for each attribute were evaluated by ANOVA and Tukey's test ($P > 0.05$). Time-intensity curves were constructed in Microsoft Excel 2010 using the parameters collected on the computer.

The results of the physicochemical analyses were analysed by univariate statistical analysis (ANOVA), and the means were compared by Tukey's test (at 5% significance). A completely randomised experimental design was used.

The software Statistical Analysis System was used (SAS, 2012).

Results and discussion

Time-intensity analysis for sweetness stimulus

Sweetness was the first attribute evaluated by the twelve assessors previously selected and trained. Table 3 shows the mean values of the four parameters evaluated in relation to sweetness, using the time-intensity analysis. No significant differences ($P > 0.05$) were observed in the sweetness time profile of the sugar-free bittersweet chocolates when compared to the sugar-sweetened chocolate. Palazzo & Bolini (2014) evaluated sweetness of milk chocolate formulated with different sweeteners, using the time-intensity analysis, and found significant differences between the samples.

Rebaudioside A is responsible for the sweet taste of stevia (Wang *et al.*, 2016). Therefore, a higher sweetness intensity and longer duration of the stimulus were expected for the samples sweetened with stevia containing 97% rebaudioside A, which was not observed in this study. This finding may be due to the differences between the contents of this glycoside that were not perceived by the assessors, once the food matrix has a strong and characteristic flavour. Bittersweet chocolate has a predominant flavour of cocoa liquor, with hints of nuts and caramel (Liu *et al.*, 2015). Dutra & Bolini (2014) have also found no differences in the sweetness profile of acerola juice sweetened with stevia containing different rebaudioside A levels, probably due to the high acidity of that juice, which diminished the perception of sweetness.

The results in Table 3 demonstrate that the fat reduction in the samples (Stevia 60% Reb A*, Stevia 80% Reb A*, Stevia 97% Reb A*) did not affect the

Samples	Tlmax	lmax	Area	Ttot
Sucrose	22.1 ^a ± 1.23	6.61 ^a ± 0.89	206.01 ^{ab} ± 8.21	56.28 ^a ± 3.13
Stevia 60% Reb A	21.8 ^a ± 2.17	6.26 ^{ab} ± 0.95	212.87 ^a ± 11.12	62.6 ^a ± 3.54
Stevia 80% Reb A	22.49 ^a ± 2.82	5.86 ^{ab} ± 1.11	176.47 ^{ab} ± 7.53	60.59 ^a ± 4.17
Stevia 97% Reb A	20.96 ^a ± 1.86	6.49 ^a ± 1.01	202.3 ^{ab} ± 10.67	60.09 ^a ± 3.22
Stevia 60% Reb A*	21.45 ^a ± 1.55	5.81 ^{ab} ± 0.79	171.05 ^b ± 7.22	57.2 ^a ± 2.96
Stevia 80% Reb A*	21.58 ^a ± 2.34	6.28 ^{ab} ± 1.22	197.41 ^{ab} ± 8.19	58.67 ^a ± 3.1
Stevia 97% Reb A*	21.96 ^a ± 2.41	5.87 ^{ab} ± 1.08	193.33 ^{ab} ± 9.25	59.64 ^a ± 2.79

Means with the same superscript letter are not significantly different at a 5% level.

*Low-fat samples.

Table 3 Time-intensity analysis for sweetness stimulus

perception of sweetness by the assessors, which was also observed in a study with low-fat ice cream (Cadena *et al.*, 2012). According to Wiet *et al.* (1993), fat reduction may cause a slight increase in sweetening power of the sweeteners used to replace sucrose. However, according to those authors, small differences in the perception of sweetness as a function of fat content are dependent on the food matrix, and thus, it cannot be generalised.

Time-intensity analysis for bitterness stimulus

Bitterness was the second attribute evaluated by the assessors. The polyphenolic compounds from cocoa confer bitter and astringent characteristics on bitter-sweet chocolates (Harwood *et al.*, 2013). Therefore, bitterness was expected in all samples due to the presence of cocoa liquor to the formulations, with a higher intensity in the samples sweetened with stevia containing lower rebaudioside A levels which contain higher stevioside levels, responsible for the bitter taste (Torri *et al.*, 2016).

As shown in Table 4, there was no significant difference ($P > 0.05$) among the seven samples for the TImax parameter. In contrast, a significant difference was observed in the Imax and Area among the sugar-free samples (Stevia 60%, 80% and 97% Reb A) and the sugar-free, low-fat samples (Stevia 60%, 80% and 97% Reb A*). According to Guinard & Mazzucchelli (1999), low-fat chocolates tend to be more bitter, as fat has the ability to mask the bitter compounds. In this study, bitterness increased with fat reduction.

A significant difference in the total duration of the stimulus was observed for all stevia-sweetened samples when compared to the sugar-sweetened sample, due to the residual bitterness characteristic of stevia (Chranioti *et al.*, 2016). Morais *et al.* (2014) analysed the TI profile in chocolate dairy dessert in the traditional version and with prebiotic and light version (sweetened with different sweeteners) and did not observe changes in the total duration of the bitterness for stevia-sweetened sample when compared to the sugar-sweetened sample.

Again, there was no significant difference ($P > 0.05$) in the temporal perception of bitterness between stevia with different levels of rebaudioside A. Bittersweet chocolate is a product with characteristic bitterness, which may have affected the perception by the assessors.

Time-intensity analysis for melting rate stimulus

Melting rate was the last attribute evaluated by the assessors. This evaluation aimed to verify possible changes in the melting rate due to the reduction in cocoa butter, and the effectiveness of inulin as a fat substitute in this product. Melting of chocolates in the mouth is defined by the characteristics of the fat phase and affects the perception of chocolate taste and texture; thus, it is considered a quality attribute (Afoakwa *et al.*, 2007).

As shown in Table 5, no significant differences ($P > 0.05$) were observed among the samples for the parameter TImax. However, the low-fat chocolates (Stevia 60%, 80% and 97% Reb A*) were significantly different from all other samples for the parameters Imax, Area and Ttot, demonstrating that there was a change in the temporal perception of the melting rate of the low-fat samples, even those containing a higher amount of inulin, which is a functional ingredient widely used to replace sugar and fat (Aidoo *et al.*, 2017).

Aidoo *et al.* (2015) studied sugar-free chocolates with prebiotic fibres and also observed changes in the melting behaviour.

Multiple time-intensity analysis (MTIA)

MTIA should be applied in studies on the development of new formulations, especially those using sugar and fat substitutes, as it demonstrates that the use of different ingredients can lead to changes in the time-intensity profile during consumption for each attribute and product, which is not assessed by the classical descriptive sensory analysis (Palazzo & Bolini, 2014).

Table 4 Time-intensity analysis for bitterness stimulus

Samples	TImax	Imax	Area	Ttot
Sucrose	17.14 ^a ± 1.09	4.47 ^c ± 0.83	111.33 ^c ± 10.1	46.98 ^b ± 4.12
Stevia 60% Reb A	19.24 ^a ± 1.17	5.89 ^b ± 0.67	174.92 ^b ± 11.87	54.12 ^a ± 3.89
Stevia 80% Reb A	19.3 ^a ± 2.06	5.78 ^b ± 0.75	176.57 ^b ± 9.93	56.54 ^a ± 3.33
Stevia 97% Reb A	18.97 ^a ± 1.36	5.81 ^b ± 0.73	175.04 ^b ± 12.34	57.74 ^a ± 4.01
Stevia 60% Reb A*	19.29 ^a ± 2.01	6.77 ^a ± 1.01	216.93 ^a ± 10.73	58.62 ^a ± 3.76
Stevia 80% Reb A*	18.55 ^a ± 2.22	7.01 ^a ± 0.98	217.51 ^a ± 11.21	56.34 ^a ± 3.91
Stevia 97% Reb A*	18.42 ^a ± 2.31	6.78 ^a ± 0.76	220.08 ^a ± 10.11	57.32 ^a ± 2.98

Means with the same superscript letter are not significantly different at a 5% level.

*Low-fat samples.

Table 5 Time-intensity analysis for melting rate stimulus

Samples	T _{lmax}	I _{max}	Area	T _{tot}
Sucrose	45.85 ^a ± 3.45	7.3 ^a ± 1.22	297.34 ^a ± 14.65	78.83 ^a ± 4.11
Stevia 60% Reb A	46.2 ^a ± 3.76	7.26 ^a ± 2.02	272.26 ^{ab} ± 13.98	78.07 ^a ± 4.51
Stevia 80% Reb A	45.29 ^a ± 3.92	7.19 ^a ± 1.45	266.5 ^b ± 14.14	77.71 ^{ab} ± 5.34
Stevia 97% Reb A	47.37 ^a ± 2.74	7.21 ^a ± 1.12	287.93 ^{ab} ± 14.54	79.35 ^a ± 5.02
Stevia 60% Reb A*	45.12 ^a ± 2.98	5.68 ^b ± 1.31	209.38 ^c ± 15.1	73.31 ^{bc} ± 4.97
Stevia 80% Reb A*	45.02 ^a ± 3.1	5.52 ^b ± 1.21	212.44 ^c ± 13.79	72.75 ^c ± 4.31
Stevia 97% Reb A*	45.1 ^a ± 2.45	5.61 ^b ± 0.99	208.7 ^c ± 14.23	73.36 ^{bc} ± 6.02

Means with the same superscript letter are not significantly different at a 5% level.

*Low-fat samples.

The time-intensity curves for the three stimuli (sweetness, bitterness and melting rate) of each sample (multiple time-intensity representations) are shown in Fig. 1.

Figure 1 is a graphical form of representing the mean values of the four parameters evaluated in relation to the three stimuli. The samples without fat reduction exhibited a temporal profile more similar to the sugar-

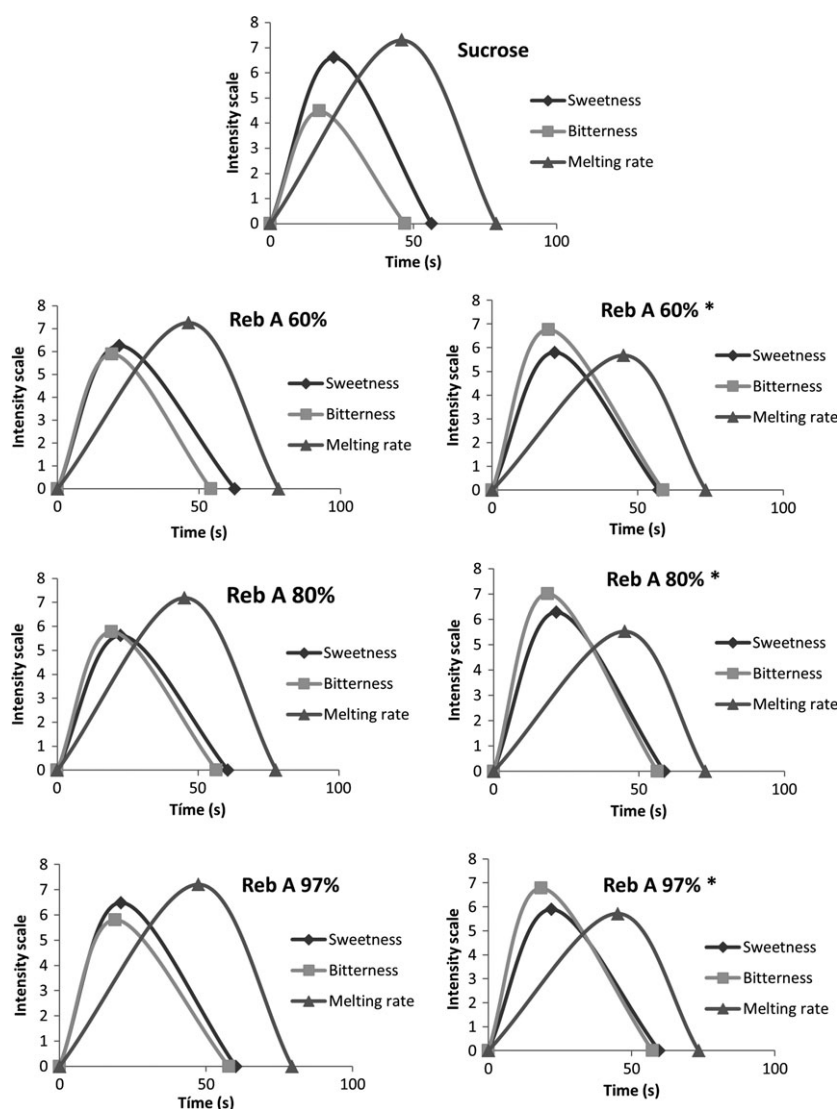


Figure 1 Multiple time-intensity analysis of semibitter chocolate samples sweetened with sucrose, with stevia 60%, 80% and 97% rebaudioside A and with stevia 60%, 80% and 97% rebaudioside A*. * Samples with reduced fat contents.

Table 6 Hardness and colour (L*, a*, b*) of bittersweet chocolate samples

Samples	Hardness (g)	L*	a*	b*
Sucrose	7180.94 ± 125.3 ^b	28.74 ± 0.17 ^b	6.96 ± 0.13 ^b	6.35 ± 0.18 ^b
Stevia 60% Reb A	6246.01 ± 137.09 ^c	28.22 ± 0.15 ^c	6.59 ± 0.22 ^c	6.03 ± 0.22 ^{bc}
Stevia 80% Reb A	6340.95 ± 104.04 ^c	28.2 ± 0.12 ^c	6.56 ± 0.15 ^c	6.07 ± 0.08 ^{bc}
Stevia 97% Reb A	6377.7 ± 116.64 ^c	28.1 ± 0.05 ^c	6.57 ± 0.08 ^c	6.03 ± 0.19 ^c
Stevia 60% Reb A*	7801.03 ± 98.08 ^a	30.08 ± 0.2 ^a	7.46 ± 0.16 ^a	7.1 ± 0.06 ^a
Stevia 80% Reb A*	7776.14 ± 121.49 ^a	30.07 ± 0.05 ^a	7.49 ± 0.18 ^a	7.11 ± 0.1 ^a
Stevia 97% Reb A*	7644.69 ± 120.38 ^a	30.05 ± 0.03 ^a	7.48 ± 0.08 ^a	7.07 ± 0.13 ^a

*Low-fat samples.

sweetened sample, mainly concerning the sweetness and melting in the mouth. The low-fat samples were more bitter and presented lower melting rate in the mouth. These results are essential for the development of products with sensory characteristics similar to the conventional product, thus affecting the consumers' purchase.

Physicochemical characterisation

Texture and colour measurements

The hardness values and colour parameters of the samples are shown in Table 6.

Changes in composition, processing and storage, among others, can affect chocolate texture, impairing its quality. Low hardness leads to the development of a sticky texture, while high hardness makes it difficult to chew (Alvis *et al.*, 2011).

The low-fat chocolates presented higher hardness values and were significantly different from the other samples. Different behaviour was observed by Rezende *et al.* (2015), who found an increase in hardness of chocolates with the increase in cocoa butter contents, as the solid particles became more dispersed with the increase in the lipid phase, causing less interference in the crystallisation process, which resulted in hard chocolates (Beckett, 2009). In contrast, Aidoo *et al.* (2017) reported that the increase in fat content also decreased the hardness of bittersweet chocolates.

In the present study, the low-fat samples had higher inulin levels, which may also have influenced the present results. Furlán *et al.* (2017) also found higher hardness in white chocolate samples containing higher inulin levels.

Regarding the colour parameters, the L* values ranged from 27.5 to 29.6, characterising the samples as bittersweet chocolates. Low-fat samples with higher inulin content had higher L*, a* and b* values. Shourideh *et al.* (2012) have shown that the increase in inulin concentration leads to an increase in L* value. Morais-Ferreira *et al.* (2016) found higher a* and b* values in low-fat food products.

Conclusion

According to the time-intensity analysis, no significant differences were observed in the sweetness temporal

profile of the six sugar-free chocolates when compared to the sugar-sweetened sample, demonstrating that stevia and inulin have proven to be excellent sucrose substitutes.

In relation to the bitterness stimulus, the low-fat samples had a more accentuated perception of this attribute by the assessors, with significant differences for I_{max} and Area when compared to the sugar-free samples.

The stimulus melting in the mouth was more affected in the low-fat samples.

Data have shown no significant differences between stevia with different rebaudioside A levels. Therefore, stevia with 60% rebaudioside A is the most indicated due to its lower cost and similar sensory profile, as demonstrated by the time-intensity analysis of sweetness, bitterness and melting rate, which is the most relevant attributes in a sugar-free, low-fat chocolate.

Although the samples with lower cocoa butter contents presented changes in bitterness and melting rate, the product may appeal to an audience who enjoys bitter chocolate and searches for light and healthier products. The sugar-free sample sweetened with stevia containing 60% rebaudioside A may be an excellent alternative healthy product similar to the conventional, once it is less caloric and sweetened with a cheap natural sweetener, besides containing prebiotic fibre with functional properties and presenting a profile similar to the sugar-sweetened sample.

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