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Freeze-dried candies from blackcurrant (*Ribes nigrum L.*) and yoghurt. Physicochemical and sensorial characterization

Diego Archaina[a,d], Natalia Sosa[a,d], Roy Rivero[a,d], Carolina Schebor[b,c,d,*]

[a] *Laboratorio de Investigación y Servicios de Productos Apícolas, Facultad de Bromatología, Universidad de Entre Ríos, Gualeguaychú, Entre Ríos, Argentina*
[b] *Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales, Departamento de Industrias, Ciudad Universitaria, Buenos Aires, Argentina*
[c] *CONICET-Universidad de Buenos Aires, Instituto de Tecnología de Alimentos y Procesos Químicos (ITAPROQ), Buenos Aires, Argentina*
[d] *CONICET, Buenos Aires, Argentina*

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ABSTRACT

The objective of this work was to develop lyophilized berry candies in order to offer a healthy alternative to traditional candies. Two formulations were developed based on blackcurrants, yoghurt and different sweeteners: honey/isomalt (F1, oriented to children); and isomalt/stevia (F2, oriented to adults). F1 showed higher water content and a_w values than F2; F1: 2.93 ± 0.19 (g H_2O/100g sample) and a_w 0.36 ± 0.06; F2: 1.79 ± 0.16g H_2O/100g sample and a_w 0.27 ± 0.06. Both formulations presented a pink color (F1: a* = 23.06 ± 3.18, b* = 4.35 ± 1.07; F2: a* = 35.42 ± 2.08, b* = 1.14 ± 0.88), however F2 lightness (41.03 ± 1.37) was much higher than that of F1 (15.31 ± 2.42). According to a qualitative sensory test, about 80% of the consumers described good attributes of the candies. A quantitative sensory test was used to describe textural attributes. The panel members identified texture changes along storage, mainly a decrease of hardness and crunchiness. The antioxidant activity input of both formulations was similar to that of kiwifruit and apple (F1: 4.53 ± 0.59 and F2: 4.24 ± 0.29 mmol Trolox/Kg), which is a positive feature, given that usually candies have reduced or null input of bioactive compounds.

1. Introduction

In recent years, the fruits with high flavonoids content have acquired considerable interest due to their potential biological and health-promoting properties. Berries represent a widely consumed group of fruits in the human diet. They are popularly consumed not only in fresh and frozen forms but also as processed and derived products, including dried and canned fruits, yoghurts, beverages, jams, and jellies (Cervenka, 2009; Nile & Park, 2014; Vagiri, Ekholm, Andersson, Johansson, & Rumpunen, 2012). Berries provide significant health benefits because of their high levels of polyphenols, antioxidants, vitamins, minerals, and fibers. The members of the genus *Ribes* are known to have very high antioxidant capacity. In this sense, there has been an increased scientific interest toward the crops belonging to this genus, not only due to their desired taste but also for the health benefits (Cervenka, 2009; Vagiri et al., 2012).

Freeze-drying is a dehydration method that has become one of the most important processes for the preservation of heat-sensitive materials and foods (Ciurzyńska & Lenart, 2011). It is interesting to note that there is a wide variety of dehydrated products obtained by freeze-drying from different berries. Lim, Tanng, and He (1995) studied freeze-dried blueberries. Franceschinis, Salvatori, Sosa, and Schebor (2014) developed a powder from blackberry juice with potential use as food colorants or healthy ingredients. Sette, Franceschinis, Schebor, and Salvatori (2016) used the freeze-drying process to obtain dehydrated raspberries, which showed a high retention of bioactive compounds. Nemzer, Vargas, Xia, Sintara, and Feng (2018) dried blueberries, tart cherries, strawberries, and cranberries by different methods, and showed that, the freeze-dried fruits showed better quality retention compared with the fruits dried with the refractance window and hot-air dried methods.

There are scarce studies on the use of berries juices, extracts or pulps as ingredients for the development of snacks or candies. Karki (2011) developed fruit leathers from different blueberry cultivars, obtaining products with high antioxidant capacity and total phenolic content. Mäkilä et al. (2014) used the blackcurrant juice press residue for the production of extruded snacks. These products showed good energy, protein and total fiber levels, but the authors emphasized about the

* Corresponding author. Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales, Departamento de Industrias, Ciudad Universitaria, Buenos Aires, Argentina.

E-mail address: cschebor@di.fcen.uba.ar (C. Schebor).

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importance to improve the sensory properties of the products in order to maintain characteristics such as taste and color of the fresh berry. Tarko, Duda-Chodak, and Semik-Szczurak (2017) developed apple chips impregnated with different berries ´extracts, obtaining high sensory scores and the enrichment in polyphenols and antioxidant activity.

The development of candies from fruits could be an opportunity to take advantage of discard fruits that do not fulfill the consumer's requirements for fresh consumption. Also, in some cases, as it happens with blackcurrant fruits, their taste is strongly acid, thus they are not frequently consumed as fresh fruits. Additionally, having new snacking options with high nutritional input could be an interesting alternative to regular candies that usually do not provide a nutritional contribution. Taking these facts into account, the hypotheses of this work consider, on one hand, that blackcurrant can be used as a fruit source to obtain high quality dehydrated candies, having an interesting input of bioactive compounds. The second hypothesis states that acceptable candies can be developed using alternative sweeteners (different from sucrose), in order to offer attractive and healthier options to the regular candies, tending to cover the demand of different consumers.

The aim of this work was to develop two different freeze-dried candies from blackcurrant fruit and perform their physicochemical and sensory characterization.

2. Materials and methods

2.1. Materials

Individual Quick Frozen (IQF) blakcurrants (*Ribes nigrum L.*) Titania cultivar from El Bolson, Argentina were used. A yoghurt starter culture (Amerex®), honey (Gala®), skimmed milk (SanCor®), isomalt (Beneo®), stevia (Dulri®), and a food grade maltodextrin DE 12 were used. Analytical-grade reagents were used in all cases.

For the texture panel several products were used as references (Table 1).

2.2. Methods

2.2.1. Development of candies

2.2.1.1. Fruit pulp preparation. IQF blackcurrants were thawed at 40 °C for 30 min and then they were milled with a mixer for 2 min, until obtaining a homogeneous pulp.

2.2.1.2. Yoghurt preparation. Yoghurt was prepared from skimmed milk, a starter culture and maltodextrin. Milk was heated at 80 °C for 15 min, and then it was cooled up to 45 °C to incorporate the starter culture (*Streptococcus thermophilus* spp. and *Lactobacillus bulgaricus* spp.) and maltodextrin (10%, added to increase the solids content). The preparation was mixed using a magnetic stirrer and placed in polyethylene flasks. The fermentation process was conducted at 40 ± 2 °C, until pH 4.5 was reached.

2.2.1.3. Formulations preparation. Two formulations were prepared by mixing the different ingredients. F1: Yoghurt (46.7 g/100 g mixture), fruit (23.3 g/100 g mixture), honey (20.0 g/100 g mixture) and isomalt (10.0 g/100 g mixture); F2: yoghurt (46.3 g/100 g mixture), fruit (23.2 g/100 g mixture), isomalt (30.0 g/100 g mixture) and stevia (0.50 g/100 g mixture).

2.2.1.4. Candies elaboration. Each formulation was molded in silicon molds and frozen for 48 h at −20 °C. Frozen candies were unmolded, put in polyethylene trays and freeze-dried. The freeze-drying process lasted 48 h and was carried out in a freeze drier Alpha 1-4 LD/2-4 LD-2 (Martin Christ, Gefriertrocknungsanlagen GmbH, Osterode, Germany). It was operated at −55 °C at a chamber pressure of 4 Pa. The obtained candies were hermetically packaged into sealed PVDC bags.

2.2.2. Physical analysis

2.2.2.1. Water content determination. The water content was determined by titration using the method proposed by Franceschinis et al. (2014).

2.2.2.2. Water activity (a_w). A_w was measured using the method proposed by Franceschinis et al. (2014).

2.2.2.3. Superficial color. Superficial color was determined by photocolorimetry using a handheld colorimeter HunterLab MiniScan EZ (Murnau, Germany). Color functions were calculated for illuminant C at 2° standard observer and in the CIELAB uniform color space. Ten replicates of each sample were analyzed.

2.2.3. Chemical analysis

2.2.3.1. Total acidity. Acidity was determined by titration using the 942.15 official method (AOAC, 2005).

2.2.3.2. pH. It was measured with a pH meter Hanna HI 2223 (Romania) using the 945.27 official method (AOAC, 2005).

2.2.3.3. Determination of total monomeric anthocyanins (TMA). TMA content was measured using the method used by Archaina, Leiva, Salvatori, and Schebor (2017). The results were expressed as mg of cyanidin-3-glucoside per 100 g of dry matter (mg cyn-3-glu/100 g d.m.)

2.2.3.4. Determination of total phenolic compounds (TPC). TPC determination was carried out using the method used by Archaina et al. (2017). Solutions of gallic acid (0–0.3 mg/mL) were used to construct the calibration curve (r^2 = 0.998). The results were expressed as mg of gallic acid equivalents per 100 g of dry matter (mg GAE/100 g d.m.).

2.2.3.5. Determination of antioxidant capacity (AC). AC was determined using the TEAC methodology used by Archaina et al. (2017). Solutions of Trolox reagent (0.02–0.12 mg/mL) were used to construct the

Table 1
Attribute definitions (Civille & Szczesniak, 1973), and standard foods used in the reference scales (Hough et al., 1994; Farroni, 2011).

Attributes	Sensory definitions	Standard foods (reference scales)
Hardness	Force required to compress a food between the molars.	Cream cheese (1), egg white (2.5), frankfurter (5), olive (6), processed cheese (7), peanut (9.5), chocolate (11), hard candy (17)
Adhesiveness to palate	Force required to remove a food adhering on the palate's surface.	Margarine (1), peach jam (3), milk Jam (6), spreading cheese (8), peanut butter (12)
Fracturability	Degree to which a food breaks, fractures or crumbles.	Pudding (1), soft sweet biscuit (2.5), craker (5), sweet biscuit (7), honey cookie (8), thin toast bread (10),DRF® candy (12), hard candy (14.5)
Cohesiveness	Degree of compression that a food experiences between the teeth before its rupture.	Pudding (1), processed cheese (5), chewing candy (8), mogul® candy (11), sugus® candy (12) chewing gum (15)
Crunchiness	Acute and quick sound perceived when a food is bitten with the incisors and the open lips.	Cereal bar (1), cracker (5), cornflake (10), honey and oat cheerios (12), thin toast bread (17)

calibration curve ($r^2 = 0.991$). The results were expressed as mmol of Trolox per Kg of dry matter (mmol Trolox/Kg d.m.)

A spectrophotometer Jenway 6505 ultraviolet–visible (Burlington, New Jersey, 157 USA), was used for TMA, TPC and AC determinations. Before measurements, candies were milled to obtain powders.

2.2.4. In vitro digestion

2.2.4.1. Simulated in vitro digestion. Oral, gastric and intestinal phases were analyzed. In all cases, samples were incubated at 37 °C in a Function Line 7000 drying stove (Heraeus, Germany) under constant stirring with an orbital shaker Vicking M-23 (Vicking SRL, Argentina) at 100 rpm. The *in vitro* digestion was carried out using the standardized method proposed by Minekus et al. (2014).

After *in vitro* digestion, the samples were centrifuged at 10,000 rpm, at 4 °C for 10 min. Finally, the soluble fraction “supernatant” and the insoluble fraction “residue” were separated and stored frozen until use.

2.2.4.2. Antioxidant capacity assay. The antioxidant capacity was measured in the fresh soluble and the lyophilized insoluble fractions after the *in vitro* digestion.

Soluble fraction: it was performed using the TEAC assay (Archaina et al., 2017).

Insoluble fraction: it was performed using the QUENCHER procedure as described by Gökmen, Serpen, and Fogliano (2009) with some modifications. One mg of lyophilized fraction was added to 10 mL of ABTS, and the mixture was vortexed for 2 min and centrifuged at 10,000 rpm for 3 min to facilitate the surface reaction with the ABTS reagent. After 30 min, the absorbance was measured at 734 nm. The same calibration curve for soluble fraction analysis was used. The results were expressed as mmol of Trolox per Kg of dry matter (mmol Trolox/Kg d.m.).

2.2.5. Sensorial evaluation

2.2.5.1. Word association task. Word association has been reported to be a quick, simple and useful qualitative methodology for exploring consumer perception of food products (Ares, Giménez, & Gámbaro, 2008). In the present study, participants were given a poll through e-mail and social networks with a photo, a short description about the blackcurrant candies, and a question (Fig. 1). All the words, descriptions and associations provided by participants were considered for the analysis. The frequency in which each association was mentioned was determined and the data analysis was performed using the Word Art web-based free service to create a words cloud. On the other hand, the associations were grouped into different categories. The grouping procedure was performed independently by three of the researchers who authored this study, considering personal interpretation of the meaning of the words. The final categories and their names were determined by consensus between the three researchers considering their three independent classifications and the discussion between them (Guerrero et al., 2010; Vidal, Ares, & Giménez, 2013). Finally, the percentage of words mentioned for the identified categories was determined.

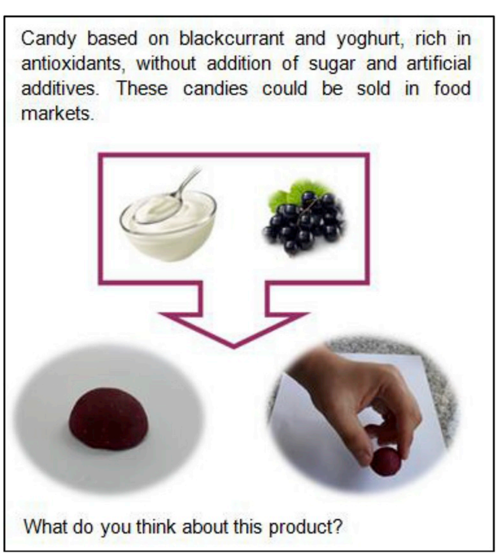

Fig. 1. Poll sent through e-mail and social networks to perform the descriptive sensorial test of the blackcurrant candy.

2.2.5.2. Texture profile panel. Sensory evaluation of the two types of candies (F1 and F2) was carried out in the sensory analysis laboratory from the Facultad de Bromatología, Universidad Nacional de Entre Ríos. In order to describe the sensory properties, the sensory profiling method was applied (ISO 13299, 2003). This method consisted of two phases, an initial phase that allowed selecting, training and validating the assessors to integrate a texture profile panel and a subsequent phase in focus on the evaluation of the samples. Twenty assessors (13 females and 7 males, 25-65 years-old) took part in this study. They were selected and recruited following international standards (ISO 8586, 2012). All of them were staff members of the Facultad de Bromatología.

a) First phase: selection, training and validation of the assessors

2.2.5.2.1. Selection phase

- *Evaluating the assessors*: selection criteria were established based on their ability to recognize five basic tastes at a time and their concentration thresholds (ISO 3972, 2011, Jellinek, 1985).

Initiation and training of assessors: solutions with specific concentrations of different substances were prepared in order to test the assessors' detection and recognition of certain odors (ISO 5496, 2006).

Ranking: samples with different concentrations of orange juice were prepared to evaluate the color attribute (ISO 8587, 2006).

Triangle test: this analysis was aimed to determine whether the assessors were able to distinguish between samples, and if so, identify differences. (ISO 4120, 2004, Meilgaard, Civille, & Carr, 1999)

Once the results from this phase were analyzed, nine members (6 females and 3 males) were selected and trained.

2.2.5.2.2. Training phase. Training took place over a period of six months with one session per week. The aim of the basic training was to develop sensory memory in each member by using specific vocabulary and scales to describe the texture of the formulated candies.

First part: a theoretical class about sensory analyses and texture attributes was dictated to instruct the selected members about texture concepts and texture profile method.

Second part: consisted in the presentation and recognition of reference scales for Argentine foods. Scales of hardness, adhesiveness to palate, fracturability and cohesiveness proposed by Hough, Contarini, and Muñoz (1994) and crunchiness scale developed by Farroni (2011) were used.

b) Second phase: evaluation of the candies samples

Once the panel was trained and validated, the assessors carried out the sensory analysis of the F1 and F2 candies stored at 25 °C for different times (fresh candies = T_0, storage for 3 months = T_3 and storage for 6 months = T_6). In this phase, 6 candies samples (F1 and F2 candies stored at 3 different times) were evaluated. Each sample in individual PVDC bags, with a 3-digit code was presented to the assessors. For each attribute, a non-structured scale with a minimum and maximum reference anchor was used. The assessors had to open the bag containing the candies and eat them to evaluate each textural attribute. Finally, they had to assess the intensity of each attribute, and then to score them in the non-structured scale.

Table 2
Physicochemical properties and superficial color of freeze-dried candies.

Property	F1	F2
Water content (g H_2O/100 S)	2.93 ± 0.19^{a}	1.79 ± 0.16^{b}
a_w (at 25 °C)	0.36 ± 0.06^{a}	0.27 ± 0.06^{b}
Total acidity (mg citric acid/100 g d.m.)	4.55 ± 0.05^{a}	4.73 ± 0.06^{b}
Total monomeric anthocyanins (mg cyn-3-glu/100 g d.m.)	39.25 ± 0.10 ^{a}	33.64 ± 0.16^{b}
Total phenolic content (mg gallic acid/100 d.m.)	53.64 ± 0.02^{a}	52.93 ± 0.17^{b}
L* value	15.31 ± 2.42^{a}	41.03 ± 1.37^{b}
a* value	23.06 ± 3.18^{a}	35.42 ± 2.08^{b}
b* value	4.35 ± 1.07^{a}	1.14 ± 0.88^{b}

Standard deviation values are included. Different letters on the rows indicate significant differences between formulations ($p < 0.05$).

Table 3
AC values (mmol Trolox/Kg candy) of the chemical extraction, and the soluble (GAR) and insoluble (Quencher) fractions obtained after *in vitro* digestion for F1 and F2 freeze-dried candies.

	Chemical extraction	GAR	Quencher
F1	4.53 ± 0.59^{a}	6.95 ± 0.47^{b}	64.68 ± 3.96^{c}
F2	4.24 ± 0.29^{a}	6.98 ± 0.64^{b}	73.67 ± 10.36^{c}

Standard deviation values are included. Different letters on each column indicate significant differences between formulations ($p < 0.05$).

2.2.6. Statistical analysis

The results were statistically analyzed by the analysis of variance (ANOVA) to determine significant differences between the samples. The analysis of the means was performed through the LSD Fisher procedure at $p < 0.05$ using the software Infostat v.2008.

3. Results

Table 2 shows some physicochemical properties studied to characterize the freeze-dried candies from blackcurrant and yoghurt. F1 and F2 showed significantly different values in all the analyzed parameters. Although the composition of the formulations was relatively similar, the sweeteners influence was relevant on the measured physicochemical properties. F1 presented higher humidity and a_w, and lower acidity values than F2. The presence of honey in F1, which is rich in highly hygroscopic low molecular weight sugars, could promote the water retention, making difficult its elimination during the freeze-drying process. In addition, honey contributed to counteract the high acidity of the blackcurrant fruit. Regarding the bioactive compounds (TMA and TPC), F1 presented slightly higher values ($p < 0.05$) than F2. It is likely that honey, besides acting as a sweetener of F1, it could have contributed with polyphenols, causing a slight raise in TPC. Concerning the superficial color, both formulations presented pink hues, however L*, a* and b* were significantly different. F2 lightness was much higher than that of F1, probably due to the higher amount of isomalt (white powder) used as sweetener in F2.

Table 4
Frequency of mention for the identified categories from the results of the qualitative sensory test performed in the freeze-dried candy.

Categories	Frequency (%)
Good aspect	46
Healthy	22
Wishes/Feeling	12
Bad aspect	11
Taste	6
Others	3

Table 3 shows the AC values of the chemical extraction and the soluble and insoluble fractions obtained after the *in vitro* digestion of F1 and F2. For each studied fraction, no significant differences were observed in the AC values between F1 and F2. Pastoriza, Delgado-Andrade, Haro, and Rufián-Henares (2011) reported AC values of the chemical extraction of kiwifruit and apples (5.2 and 4.8 mmol Trolox/Kg sample, respectively), which are similar to those obtained for the blackcurrant candies, however the portion size and the frequency of consumption of this product in comparison to fruits should be considered.

The AC values obtained for the soluble fraction by the GAR method were higher compared to the values obtained by conventional chemical extraction, indicating that upon the digestion process there was a release of bioactive compounds with antioxidant activity. Similar results were reported by Pastoriza et al. (2011) and Wootton-Beard, Moran, and Ryan (2011) for several food matrices.

Regarding the AC that remained in the insoluble fraction, Pastoriza et al. (2011) reported diverse AC values in various foods. Karas, Jakubczyk, Szymanowska, Złotek, and Zielinska (2017) reported that the AC after the *in vitro* digestion is dependent of the food matrix due to the interaction between the bioactive compounds and the other food components. The AC values for the insoluble fraction of the blackcurrant candies were higher than the values corresponding to the chemical extraction and the soluble fraction of digestion. In addition,

Fig. 2. Cloud of words obtained from the results of the qualitative sensory test performed for the freeze-dried candy.

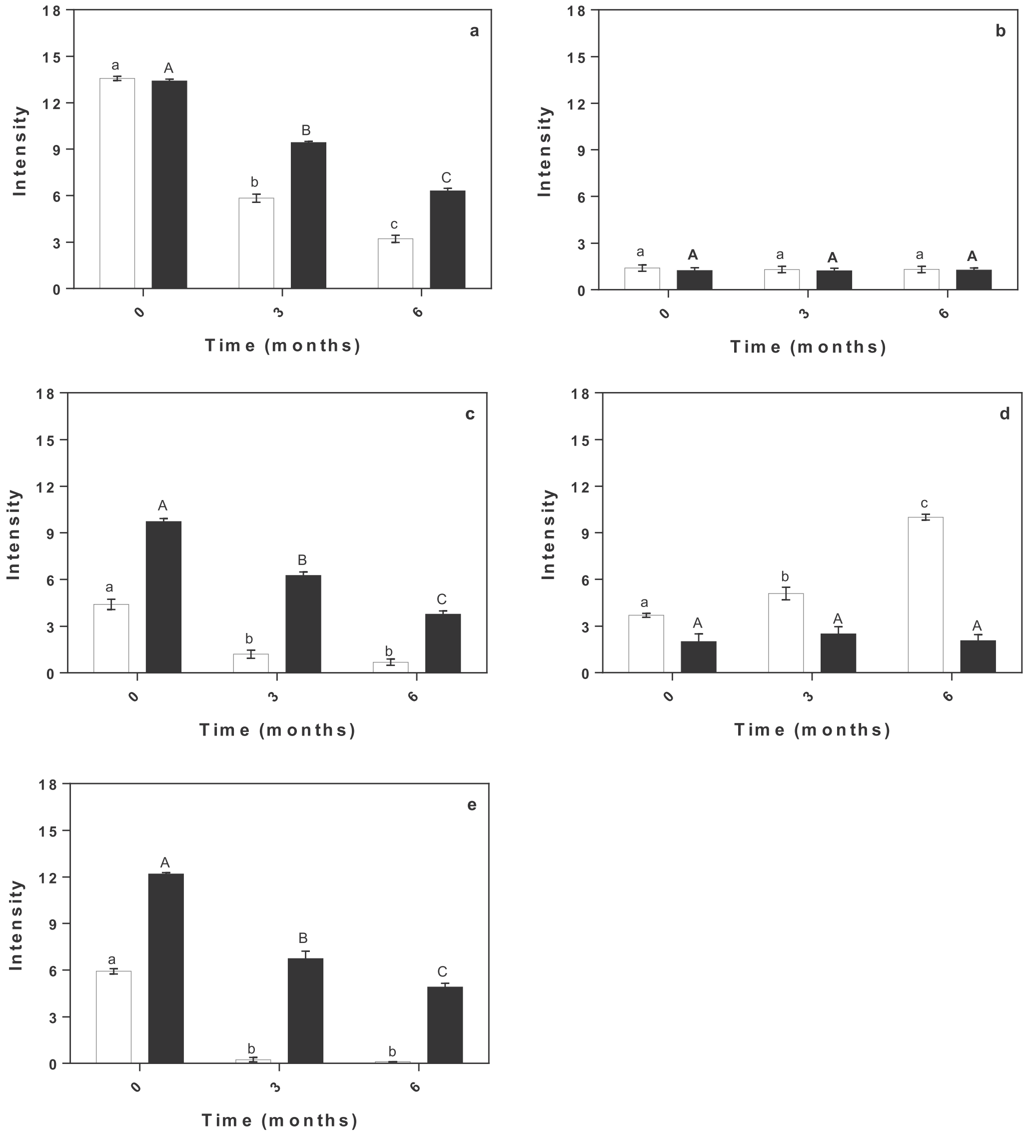

Fig. 3. Average intensity for each attribute, hardness (a), adhesiveness to palate (b), fracturability (c), cohesiveness (d), and crunchiness (e), perceived by the trained panel members along the storage time at 25 °C for F1 (white bar) and F2 (black bar) freeze-dried candies. Relative error values are included. Different letters indicate significant differences between means ($p < 0.05$). Small letters indicate significant differences for F1; while capital letters indicate significant differences for F2 at different times.

after the *in vitro* digestion, most of the candy samples belonged to the insoluble fraction (74% and 86% for F1 and F2, respectively). The gastrointestinal system is constantly exposed to reactive oxygen species (ROS) (Halliwell, Rafter, & Jenner, 2005). Therefore, the presence of antioxidants may play a role in maintaining the redox balance against different oxidants, which could prevent diseases of the gastrointestinal tract related to the generation of ROS during digestion processes (Leopoldini, Chiodo, Russo, & Toscano, 2011).

Sensory studies were carried out in order to: a) explore consumer's perception of our products, and b) analyze several textural parameters with a trained panel along the storage time at 25 °C. Fig. 2 shows a cloud of words with the results obtained in the qualitative sensory test. A total of 222 volunteers participated in the test, and 367 words were obtained. The participants showed positive and negative associations about the blackcurrant candy. The associations mentioned by the higher percentage of participants were *interesting* (n = 45), *very good idea* (n = 45), and *healthy* (n = 39); while the negative associations such as *bad aspect* (n = 5), *disgust* (n = 3) and *strange* (n = 2) were mentioned by the lower percentage of participants. Table 4 shows the percentage of mention for the identified categories. Six categories were obtained by consensus among the three researchers that participated in the data analysis process. The percentage of consumers that mentioned the category *Good aspect* (46%) was higher compared to the rest, while the category with the lowest percentage was *Others* (3%) including words such as *children*, *snack*, *school*, among others. This study indicates that the freeze-dried blackcurrant candies can have a good acceptance by the consumers.

Fig. 3 shows the average intensity for several textural attributes perceived by the trained panel members. Regarding the hardness attribute (Fig. 3a), it presented an average value of 12, similar to chocolate (value = 11) of the reference scale for both candies (t = 0), nevertheless the hardness value decreased significantly with storage time. A higher decrease of the hardness values was observed for F1, being perceived similar to egg white (value = 2.5) at 6 months storage, while at the same storage time F2 was perceived similar to olive (value = 6 of the reference scale). The adhesiveness to palate values (Fig. 3b) were similar to margarine (value = 1) of the reference scale. These values did not show significant differences, being practically constant over time for both samples. In the case of the fracturability attribute (Fig. 3c), F2 presented higher values than F1 at all the studied times. Also, this variable showed a significant decrease with storage time for both candies. The honey used in F1 produced a compact structure, while in the candy sweetened with isomalt, a porous and fragile structure was generated. The cohesiveness values (Fig. 3d) increased significantly along time for F1, while those values remained constant for F2. This would indicate a gumminess increase for F1, requiring more compression before breaking. Finally, Fig. 3e shows the crunchiness values. Similar values to cracker (value = 5) and honey and oat cheerios (value = 12) were registered for the fresh F1 and F2 candies, respectively. A significant decrease in the intensity of the crunchiness attribute was observed along storage time, and F1 lost this attribute within the first 3 months at 25 °C, turning the candy soft and chewable.

4. Conclusions

The development of freeze-dried candies using blackcurrant as a fruit source, and different sweeteners, in order to reduce the consumption of sugar, was successfully tested. This work shows that it is possible to add value to the blackcurrant fruit, which is consumed very little as fresh fruit due to its high acidity, and also to sub-standard fruits that are not sold for direct consumption. Additionally, alternative sweeteners allowed obtaining two different formulations, that showed relatively similar functional properties, but very different textural characteristics. F1 was intended for children consumption, then, it was sweetened with honey. F2 could be oriented to adults, containing low calories sweeteners as isomalt and stevia. The antioxidant activity of both candies formulations (in a range between 4.2 and 4.5 mmol Trolox/Kg candy) was similar to that of fruits like kiwifruit and apple; this is a positive aspect if it is considered that conventional candies have poor or null functional properties. Upon *in vitro* digestion a high AC was retained in the insoluble fraction (in a range between 65 and 74mmolTrolox/Kg candy), suggesting a relevant contribution to the total antioxidant capacity as the non-extractable antioxidants could produce a positive biological effect on the intestinal microbiota.

A qualitative sensory test indicated that the developed candies were accepted by about 80% of the public, suggesting that the local population could be interested in this new type of candies, not only due to their appearance characteristics, but also due to their nutritional input.

The texture profile evaluation showed that the candies suffered changes in their attributes along the storage time (although water content did not change along storage), suggesting that further studies are needed in order to determine the cause of these changes and establish the sensory shelf life.

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