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Different methods for textural evaluation of freeze-dried candies during storage.

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Practical applications

The texture is a complex attribute of the food matrices and its changes are decisive for the acceptance of consumers. Its description by one unique technique may be very difficult, for this reason in this work, we showed that the joint use of different methods like instrumental, sensory and image techniques would allow studying this attribute and its evolution along time in an integral way.

Abstract

The textural changes along storage of two freeze-dried candies developed from blackcurrant fruit, unflavored yogurt and different alternative sweeteners: one of them sweetened with honey/isomalt (HI) and another one sweetened with isomalt/stevia (IS), were analyzed through three different methods (instrumental, sensory and image analysis). Fresh candies were in the supercooled state, and presented different structural and textural characteristics (HI: compact and homogeneous, and IS: porous and crunchy), with Fmax values of 139 ±14 N and 174 ± 16 N for HI and IS, respectively. After storage, the instrumental analysis showed aprox. 60% average drop in F_{max} and W values, in agreement with the decrease observed by sensory analysis in hardness, fracturability and crispness attributes. Image analysis showed an increase in the parameters related to the homogeneity and the uniformity/smoothness for HI. A Pearson's Correlation Coefficients analysis showed that there was a good correlation between the three used techniques, suggesting that the joint use of these methods could be performed for a better understanding of complex food texture.

Keywords

Blackcurrant, texture, instrumental test, sensory study, image analysis

1. Introduction

Texture is a cognitive property assigned to foods based on how senses interact with the food by vision, touch, and oral processing, and involves the rheological and structural attributes of the food products (Chen & Opara, 2013; Pascua et al., 2013). In this sense, is it possible to carry out a complete characterization of the texture of food? Generally, this is very complex; however, the set of sensations that make up the texture can be comprehensively evaluated using different techniques such as instrumental methods, sensory analysis and image analysis (Anton and Luciano, 2007).

Instrumental analysis, using rheological or force-deformation tests, are the most widely used methods to evaluate the mechanical properties related to texture of foods (Lu & Abbott, 2004; Liu, 2019).

Alternatively, sensory analysis allows evaluating the food properties by using standardized techniques that are based on human perception with one or more of their senses. It is important to use trained sensory panels for objective evaluation of different sensory food properties (Pematilleke et al., 2022). On the other hand, image analysis, using electron scanning microscopy (SEM) and gray level co-occurrence matrix (GLCM) has become a powerful tool to study and predict changes in the textural characteristics of foods (Zheng et al., 2006; Pieniazek & Messina, 2017). In this case, SEM microscopy is a good alternative for image acquisition while the GLMC matrix allows obtaining the following textural parameters: contrast (CON), correlation (COR), entropy (ENT), energy (ASM) and homogeneity (HOM). Nouri et al. (2018) reported that the GLCM texture features showed high correlations with the instrumental texture parameters and physicochemical properties in bread during storage. Additionally, Roa Andino et al. (2018) studied freeze-dried bananas and showed that there was a good correlation between the image analysis and instrumental techniques.

In this work two formulations of freeze-dried candies containing blackcurrant fruit, yogurt and alternative sweeteners, and having different textural characteristics, were used as freeze-dried candy model systems to evaluate texture changes during storage. The existing studies concerning texture of freeze-dried sweet snacks are scarce. Regarding instrumental methods, Silva-Espinoza et al. (2021) used a penetration test to evaluate some textural properties of freeze-dried orange puree snacks. Additionally, Ciurzyńska et al. (2022) studied the mechanical properties of freeze-dried fruit snacks using a cutting test. Considering sensory studies with trained panels, Wojdyło et al. (2016) carried out the evaluation of different textural attributes of freeze-dried jujube fruits, and Kidoń and Grabowska (2021) evaluated the hardness and the crispness of freeze-dried red-fleshed apple cubes. Regarding image analysis studies, Pieniazek and Messina (2017, 2018) employed the SEM microscopy and the GLMC matrix for the image texture analysis of freeze-dried snacks from peach and banana, respectively.

The objective of this work was to make a comprehensive texture analysis of freeze-dried fruit candies during storage. The use of three different methodologies, addressing both macro and micro structural changes of the material, offers a new integral insight, considering the complexity and diversity of textural features that can evolve during food storage. This approach is particularly valuable for processed candies having different sweeteners that influence the evolution of the mechanical characteristics during storage, and thus their stability.

2. Materials and Methods

2.1. Development of freeze-dried candies

Two different candies from blackcurrant fruit, unflavored yogurt and different sweeteners were prepared. Frozen blackcurrants (*Ribes nigrum*) Titania cultivar (El Bolson, Argentina) were used. Yogurt was prepared with a starter culture (Amerex®), following the procedure described by Archaina et al. (2019). Regarding the sweeteners, honey (Gala, Argentina), isomalt (Beneo, Germany), and stevia (Dulri, Argentina) were employed.

In order to define the final proportion of the ingredients, an internal sensory panel composed of 15 persons who evaluated different preliminary formulations was used

(Watts et al.,1992; Moskowitz et al., 2012). Blackcurrant fruits were strongly acid and not sweet, therefore several ingredients were added to overcome these negative flavor aspects and develop sensory acceptable candies. Yogurt was added to soften the acid taste. Regarding sweetness, a low calories candy option was developed with the addition of stevia, however the aftertaste was rejected by the panel, so a mixture of isomalt and stevia was selected. Additionally, isomalt contributed to get better texture features, and reduce hygroscopicity upon freeze-drying, fact that was also considered for the other candy option, which was sweetened with a mixture of honey and isomalt.

The formulation corresponding to the candy sweetened with honey/isomalt (HI) was as follows: yogurt (46.7 g/100g), fruit pulp obtained by crushing the fruit (23.3 g/100g), honey (20.0 g/100g), and isomalt (10.0 g/100g). In the case of the candy sweetened with isomalt/stevia (IS), the ingredients proportion was as follows: yogurt (46.3 g/100g), fruit pulp (23.2 g/100g), isomalt (30.0 g/100g) and stevia (0.50 g/100g). The obtained formulations were molded in silicon molds with hemisphere shape: 2.20 cm diameter, and 1.90 cm height and frozen during 48h. The frozen candies were unmolded and freeze-dried for 48h in a freeze-drier Alpha 1-4 LD/2-4 (Martin Christ, Gefriertrocknungsanlagen GmbH, Osterode, Germany), which operated at -55°C and a chamber pressure of 4 Pa.

2.2. Methods

2.2.1. Thermal transitions analysis

The glass transition temperature (T_g) was measured using a DSC 822e Mettler Toledo calorimeter (Schwerzenbach, Switzerland) using the procedure described in Archaina et al. (2019).

2.2.2. Water content analysis

The water content was determined by Karl Fisher titration with a Karl Fisher TIM 980 titration manager (Radiometer Analytical, France) at 25°C.

2.2.3. Textural analysis

Candy samples were packed into hermetically sealed PVCD bags, coated with aluminum foil. Then they were stored in an incubator HCP108 (Memmert GmbH, Schwabach, Germany) at 25 ± 2°C, at a constant relative humidity (RH) of 60%.Texture analysis was carried out at the beginning (t=0), after 3 months (t=3) and after 6 months (t=6) of storage.

2.2.3.1. Instrumental method

The instrumental texture was analyzed by a puncture test using a Universal Testing Machine Instron 3342 (Massachusetts, USA) connected by a computer to Instron Bluehill Material Testing software. A 3 mm diameter spherical stainless steel probe was used. The test ended when each candy sample was completely pierced. The operational conditions used were: head speed= 0.5 mm/s, time interval (data capture)= 100 ms, load range= 500 N. From the force-deformation curves, the following parameters were obtained: maximum force (F_{max}), distance corresponding to the maximum force (ΔF_{max}), and fracture work (W) (Sette et al., 2016).

2.2.3.2. Sensory method

To describe the sensory texture of both freeze-dried candies, a sensory profiling method following the procedure described in Archaina et al. (2019) was applied.
In the selection phase twenty assessors (25-65 year-old) participated and the following tests were performed: a) basic tastes recognition (Jellinek, 1985; ISO 3972, 2011); b) detection and recognition of odors (ISO 5496, 2006); c) color attribute evaluation (ISO 8587, 2006); d) difference between samples – triangle test (Meilgaard et al., 1999; ISO 4120, 2004).
From the results obtained in the selection phase, nine assessors (6 females and 3 males) were selected and trained. In the training phase different attributes were

evaluated, and scales with different reference foods for hardness, adhesiveness to palate, fracturability and cohesiveness (Hough & Contarini, 1994), and crunchiness (Farroni, 2011) were used (**Table 1**).

After six months of training, the trained judges carried out the evaluation of the HI and IS candies stored at 25ºC at the different chosen times. For each evaluated attribute the judges used a non-structured scale with a minimum and maximum reference anchor (Cruz et al., 2003; Philippe et al., 2004; Tronchoni et al., 2018).

[insert Table 1.]

2.2.3.3. Image analysis method

Images acquisition

The acquisition of images of the freeze-dried candies was carried out by scanning electron microscopy (SEM) using a microscope (Supra 40, Zeiss, Germany). Samples were cross-sectioned near the surface using a scalpel, the cut was always performed in the same direction. The pieces were mounted on an aluminum support using a double-sided conductive carbon adhesive tape, and then were coated with gold nanoparticles using a sputter coater (Cressington Scientific Instruments). The images were taken with the detector within the lens, using an acceleration voltage of 3.00 kV. For each sample (t=0, t=3 and t=6), ten images at magnification of 250X were obtained, and they were stored as bitmaps in a gray scale with brightness values between 0 and 255 for each pixel constituting the image (Pieniazek et al., 2018; Roa Andino et al., 2018).

Images analysis

Textural parameters were calculated from SEM images using the Gray Level Co-occurrence Matrix method (GLCM) and MATLAB 8.4 software (The Math Works Inc., Massachusetts, USA) as described by Pieniazek et al. (2018). The size of each sample (region of interest: 122 x 122 pixels) was the same for all the evaluated images. The textural parameters contrast (CON) **equation (1)**, entropy (ENT) **equation (2)**, energy (ASM) **equation (3),** and homogeneity (HOM) **equation (4)** were calculated as follows:

$$CON=\sum_{i=0}^{n-1}\sum_{j=0}^{n-1}(i-j)^2\,Pd,\Theta(i,j) \quad \textbf{(1)}$$

$$ENT=\sum_{i=0}^{n-1}\sum_{j=0}^{n-1}Pd,\Theta(i,j)^2\,\mathrm{Log}\,P(i,j) \quad \textbf{(2)}$$

$$ASM=\sum_{i=0}^{n-1}\sum_{j=0}^{n-1}Pd,\Theta(i,j)^2 \quad \textbf{(3)}$$

$$HOM=\sum_{i=0}^{n-1}\sum_{j=0}^{n-1}\frac{Pd,\Theta(i,j)}{1+(i-j)} \quad \textbf{(4)}$$

where:

P= gray level co-ocurrence probability of i and j

d= co-ocurrence distance between i and j

Ɵ= co-ocurrence direction between i and j

i= reference pixel

j= neighboring pixel

2.3. Statistical analysis

All statistical analyses were performed using the software Infostat v.2008 (Di Rienzo et al., 2008). From the results, an analysis of variance (ANOVA) was performed and differences between means were established by using the LSD Fisher procedure. The textural parameters obtained by instrumental, sensory, and image analyses were compared descriptively with a principal component analysis (PCA). Furthermore, a correlation was established between them through a study of Pearson's correlation coefficients (PCC).

3. Results

3.1. Textural properties stability

Textural characteristics have great relevance in food products, and the knowledge of the evolution of texture attributes along storage is a requisite to establish the product´s shelf-life. In this sense, with the purpose of studying the changes produced on the texture of freeze-dried candies during storage (initial time (t=0), 3 months (t=3) and 6 months (t=6)), an instrumental puncture test, a sensory test with a trained panel, and an image analysis using scanning electron microscopy (SEM) and gray level co-existence matrix (GLCM) were performed.

The candies selected for this study presented a relatively low water content (2.9 $\pm$ 0.2 and 1.8 $\pm$ 0.2 g H_2O/100 g candy, for HI and IS, respectively), which remained constant during storage. However, due to the low glass transition temperature (T_g) of many of the ingredients used in each formulation (i.e.: fruit pulp, honey), both candies were in the supercooled state at 25°C. The T_g values were 8ºC, and 24°C for HI and IS, respectively, suggesting that changes in texture could be expected along time (Sosa y col., 2012), particularly for HI because of the lower T_g value.

3.1.1. Instrumental analysis

Figure 1 shows the characteristic force-distance curves for HI and IS candies, obtained at 25ºC through a puncture test at different storage times, while **Table 2** shows the mechanical parameters obtained from the force-displacement curves. F_{max} parameter is related to sample firmness or hardness, ΔF_{max} is the distance corresponding to the maximum force and is related to sample deformation before breaking. W is a measure of the energy necessary for fracture. High values of F_{max}

and W parameters indicate a higher expression of the textural characteristic associated with them, while for the ΔF_{max} parameter, low values are related to a higher brittleness and high values are indicative of a higher gumminess (Sette et al., 2016).

Regarding HI candies (**Figure 1A**), similar symmetric and rounded curves with higher maximum penetration force values at t=0 and t=3 were obtained. In IS samples (**Figura 1B**), the first fracture event takes place earlier, at the beginning as well as at the end of the storage time, which denotes a more brittle material when compared to HI candies. On the other hand, a succession of several peaks is observed with abrupt drops in force related to a rapid propagation of the fracture, this corresponding to a porous and crispy structure typical of freeze-dried products (Prothon et al., 2003). The maximum peak (F_{max}) can be ascribed to the main structural breakdown of this type of structure. The significantly lower fracture work of IS samples at the end of the storage is mainly related with lower force events all over the test, including that of F_{max} (Uscanga et al., 2020). The absence of jaggedness in IH curves **(Figure 1A)** with a unique rounded peak evidences a rubbery and homogenous material (Martínez-Navarrete et al., 2019). The honey addition to the formulation yielded in these samples a more compact structure that can support a greater deformation ($>\Delta F_{max}$) before breaking (Castro Montero, 2007), while maintaining a certain firmness or hardness degree at least until 3 months of storage.

In general, it can be observed that both candies showed substantial changes in mechanical properties along storage. Maximum force for the major fracture drastically decreased after six months, curves smoothed out, and samples became more deformable, requiring less energy to reach their breaking point, a fact that could be associated with a loss of the original structural properties of both candies.

[insert Figure 1.]

[insert Table 2.]

3.1.2. Sensory study with a trained panel

Table 3 shows the values perceived by the trained panel judges for each attribute at the different storage times, and the corresponding food of the used reference scale. Both formulations showed changes in all the studied attributes, which determined important differences in their textural properties. Fresh HI (t=0) was hard like a chocolate and slightly crunchy like a cracker, however after 6 months of storage it was soft and chewy showing low values of hardness (similar to a hard-boiled egg white) and crunchiness (similar to a cereal bar), and presented a high value of cohesiveness (similar to a raisin). On the other hand, fresh IS (t=0) was very hard like a hard candy, fragile like a thin toast bread, and crunchy like honey and oat cheerios, and after storage it lost completely its textural properties (t=6). Regarding the adhesiveness attribute, it remained constant throughout the storage in both candies, with values in the range of 1.22 and 1.40, like margarine.

[insert Table 3.]

3.1.3. Images study

Figure 2 shows SEM micrographs at 250 X of cross sections of HI (**Figure 2A**) and IS (**Figure 2B**) candies stored during 0, 3 and 6 months. Both fresh candies (t=0) presented a disorganized and irregular microstructure, but with substantially different porosity degree. This is in accordance with the results reported by Mousavi et al. (2007), who established that during the freezing stage of the freeze-drying process the growth of ice crystals breaks, pushes and compresses the material, which leads to the formation of pores and cavities once the ice is sublimated. The observed microstructure was very different for the two formulations, which could be related to the different sweeteners used for each type of candy, that might have affected their physical state.

The micrographs for HI (**Figure 2A**) showed that the microstructure of the candy became more uniform when increasing time; in the candies stored for 3 months (t=3) a considerable reduction in the amount of pores was observed, while in those stored for 6 months (t=6) no pores were observed. This behavior is in accordance with the fact that HI candies were in the supercooled state at 25°C ($T-T_g$= 17°C), then physical changes like collapse and pores reduction can be expected (Levine & Slade, 1992). In the case of IS (**Figure 2B**), although a pore decrease was also detected, the changes were not as marked through the visual analysis of the SEM images. As expected according to the T_g values, more severe changes were detected for HI candies. Usually relevant changes are not expected for samples having T_g values very close to the storage temperature, as is the case of IS, which showed significant changes in mechanical properties, but kept certain crispness along storage, probably due to the retention of some characteristics at the microstructural level, like porosity. Roudaut et al. (1998) informed that in some cases, texture features do not follow the expected behavior in relation to T_g. These authors suggested that the occurrence of sub-T_g relaxations may happen at ($T-T_g$) values close to zero, inducing changes on the material texture.

[insert Figure 2.]

Table 4 shows image texture values for freeze-dried candies obtained from SEM micrographs at 250 X. The textural parameters calculated from the GLCM matrix are used to describe the SEM image and can be related to the textural characteristics of the sample. The CON parameter is related to hardness, the ENT parameter is used to characterize the roughness, the ASM parameter shows the homogeneity, and the HOM parameter measures the uniformity/smoothness. High values of these parameters indicate a higher expression of the textural characteristic associated with them (Pieniazek & Messina, 2016).

For both candies a significant decrease in their CON values was observed through storage time, suggesting that their hardness diminished. When comparing both formulations, CON values obtained for IS were higher than those for HI, which would indicate that the candies sweetened with isomalt and stevia showed a higher hardness along the storage study. ENT values did not show significant differences during the storage for each candy. Regarding the homogeneity, for HI, the ASM values increased significantly, which could indicate that the candies sweetened with honey and isomalt became smoother along time, while for IS the ASM values did not present significant differences. This could point out that this candy remained uniform during storage. On the other hand, HOM values presented a similar trend as that observed for the ASM parameter. This may suggest that these parameters were related to each other.

Based on these results, the candy sweetened with isomalt and stevia presented higher CON and ENT values and lower ASM and HOM values than the candy sweetened with honey and isomalt. This would indicate that, in general, the IS candy microstructure is harder and rougher and at the same time less homogeneous and uniform than that of HI candy.

[insert Table 4.]

3.2. Principal Components Analysis (PCA)

A Principal Component Analysis (PCA) of the textural properties of the freeze-dried candies measured by the three different methods is shown in **Figure 3**. As the used methods did not have the same scales, a previous normalization of the data was performed (Jayalakshmi & Santhakumaran, 2011).

The two principal components (CP1 and CP2) explained 87.0% of the variability of the data with a correlation coefficient of 0.98. The CP1 explained 69.6% of the data variability, with the following correlation coefficients for each attribute: instrumental hardness (0.81), sensory hardness (0.98), sensory fracturability (0.96), sensory cohesiveness (0.84), sensory crunchiness (0.92), CON (1.00), ASM (0.95), and ΔF_{max} (0.78). The variability of parameters W and ENT was mainly explained by CP2 with a correlation coefficient of 0.83 and 0.75, respectively.

At t=0 both HI and IS candies were grouped on the right side of the graph, showing that their textural properties were characterized by the hardness (explained by the three used methods), the fracturability and the crunchiness. For the specified attributes, IS presented higher values than HI.

Regarding HI, the samples moved to the left side of the graph along storage time, and the textural characteristics at t=6 became governed by the ASM, HOM, ΔF_{max}, and cohesiveness. It is interesting to note that these attributes were in the opposite direction to those that predominated in the sample at t=0, indicating that the candy lost its high hardness, fracturability and crunchiness to become rubberier, homogeneous, uniform and smooth, and experienced a higher degree of deformability (ΔF_{max}); accentuating these characteristics in the sample corresponding to t=6. The loss of hardness was in agreement with the decrease in F_{max} (**Figure 1A**) and W (**Table 2**) in the instrumental test, the decrease in the perception of this attribute by trained judges (**Table 3**) and the decrease in the value of the parameter CON in the images test (**Table 4**). In addition, the candy became softer and chewier, which was closely related to the increase in cohesiveness and the decrease in crunchiness (**Table 3**) as described by the judges of the texture sensory panel, and also with the increase of the AMS and HOM textural parameters in the image test (**Table 4**).

Meanwhile, IS experienced a shift to the left side of the graph as well, but at a lower intensity than that observed for HI. At t=3, the hardness, fracturability and crunchiness were almost completely lost, which was reflected in a noticeable decrease in the perception of these attributes compared to the samples at t=0. These characteristics disappeared at t=6, marking a total loss of the original texture of the product. In this case, the freeze-drying process allowed obtaining a hard but porous product at t=0 (**Figure 2B**), however, these characteristics were mainly lost over time. This could be correlated with the results obtained in the instrumental test for IS, which at t=6 showed a 60% and 66% drop in the values of F_{max} and W, respectively with respect to the value at t=0 (**Figure 1B**). In the texture sensory test, the trained judges detected a considerable change too. The hardness values were 14 (t=0) and 6 (t=6), which in percentage terms represented a decrease of 65%

(**Table 3**). On the other hand, the loss of IS textural characteristics was related to the decrease in crunchiness as described by trained judges (**Table 3**).

[insert Figure 3.]

3.3. Pearson's Correlation Coefficient (PCC)

In order to establish the possible correlation between the different methods used in this work, a Pearson's Correlation Coefficients (PCC) analysis was performed. **Figure 4** shows the PCC between the different parameters obtained from the three methods used to study the texture of HI (**Figure 4A**) and IS (**Figure 4B**) freeze-dried candies.

The hardness, measured for the instrumental (IH), sensory (SH) and image (CON) methods, showed PCC values close to 1 indicating that there was a positive and direct correlation between these parameters for both freeze-dried candies. Also, the hardness, had a similar behavior in the correlation with the sensory fracturability (SF) and the sensory crunchiness (SCru) for both candies. Contrary, the hardness, measured for the three methods, showed negative PCC values with the sensory cohesiveness (SCo), the energy (ASM) and the homogeneity (HOM) parameters, indicating that when the hardness values increases, the others evolved in the opposite direction in both candies.

The SCo showed a positive and direct correlation with the ASM and HOM parameters, indicating that when cohesiveness increased, homogeneity and uniformity also increased for both candies.

Regarding to the SCru parameter, it had a negative correlation with the ASM and HOM parameters for both candies. This would indicate that when the candies loose crunchiness, their texture is more uniform and homogeneous. On the other hand, when the crunchiness of the candies decreased, their elasticity increase showed high and negative PCC values for SCru-ΔF_{max} correlation.

The W parameter had a positive correlation with F_{max} parameter, but a negative correlation with ASM and HOM parameters, showing good PCC values in all cases.

[insert Figure 4.]

4. Conclusions

As the study of textural properties is very complex in foods, this work focused on the use of three different techniques (instrumental, sensory and image methods) to analyze in depth the texture developed in freeze-dried candies and its behavior throughout storage. The obtained results suggest that the three used techniques allowed a better and complementary understanding of the mechanical behavior of the studied samples, which could be difficult to attain through a unique method. Also, statistical tools were valuable to interpret in a global way the usefulness of the three proposed techniques. In this sense, the Principal Component Analysis allowed interpreting the evolution of the texture of the two candies formulations, while the Pearson's Correlation Coefficients showed that there was a good correlation between the different techniques used.

The impact on the physical, structural and sensory properties of two different candies, and the integrity loss of their structure along storage could be attributed to the used sweeteners. In the case of the candies containing honey, a compact and homogeneous texture was obtained, while the other candies presented a porous and crunchy texture due to the influence of isomalt on the physical structure. In both cases, the textural changes produced through storage were noticeable. Instrumental and sensory analysis techniques revealed the changes produced at the macrostructural level, while the study through image analysis allowed to obtain a better visualization of the changes produced at the microstructural level. Overall, these techniques allowed distinguishing the evolution of the texture of two different formulations through 6 months' storage. This suggests that the joint use of the instrumental, sensory and image methods would permit studying in an integral way the textural characteristics and their evolution along time in complex food formulations, so they could be considered for future applications in the study of other food matrices.

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Author contributions

Diego Archaina: formal analysis (lead); data curation (lead); writing original draft, review and editing (lead). **Facundo Pieniazek:** formal analysis (supporting); data curation (supporting); methodology (supporting). **Valeria Messina:** data curation (supporting); methodology (supporting); supervision (supporting). **Daniela Salvatori:** review and editing (supporting); funding acquisition (supporting); conceptualization (lead). **Carolina Schebor:** writing – original draft, review and editing (supporting); supervision (lead); funding acquisition (lead); conceptualization (lead).

Ethical Statements

Conflict of interest: All the authors certify that they have NO affiliations with or involvement in any organization or entity with any financial interest, or non-financial interest in the subject matter or materials discussed in this manuscript.

Informed consent: Written informed consent was obtained from all study participants.

Data availability statement

The data that support the findings of this study are available on request from the corresponding author

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Figure 1: Force-distance curves for HI (**A**) and IS (**B**) candies, obtained at different storage times. For both candies: t=0 (initial time, red), t=3 (three months, black), t=6 (six months, blue).

Figure 2: SEM micrographs performed at 250 X magnification of HI (A) **and IS** (B) candies at different storage times.

Figure 3: Principal component analysis for the freeze-dried textural properties of both candies stored for different times. HI: t=0 (●), t=3 (●), t=6 (●), and IS: t=0 (○), t=3 (○), t=6 (○). **Coding**: F_{max}= instrumental hardness, SH= sensorial hardness, SF= sensorial fracturability, SCo= sensorial cohesiveness, SCru= sensorial crunchiness, CON= contrast, ENT= entropy, ASM= energy, HOM= homogeneity.

Figure 4: Pearson's correlation coefficients between the different parameters obtained from the three methods of texture analysis for HI (**A**) and IS (**B**) candies. **Coding**: F_{max}= instrumental hardness, SH= sensorial hardness, SF= sensorial fracturability, SCo= sensorial cohesiveness, SCru= sensorial crunchiness, CON= contrast, ENT= entropy, ASM= energy, HOM= homogeneity.

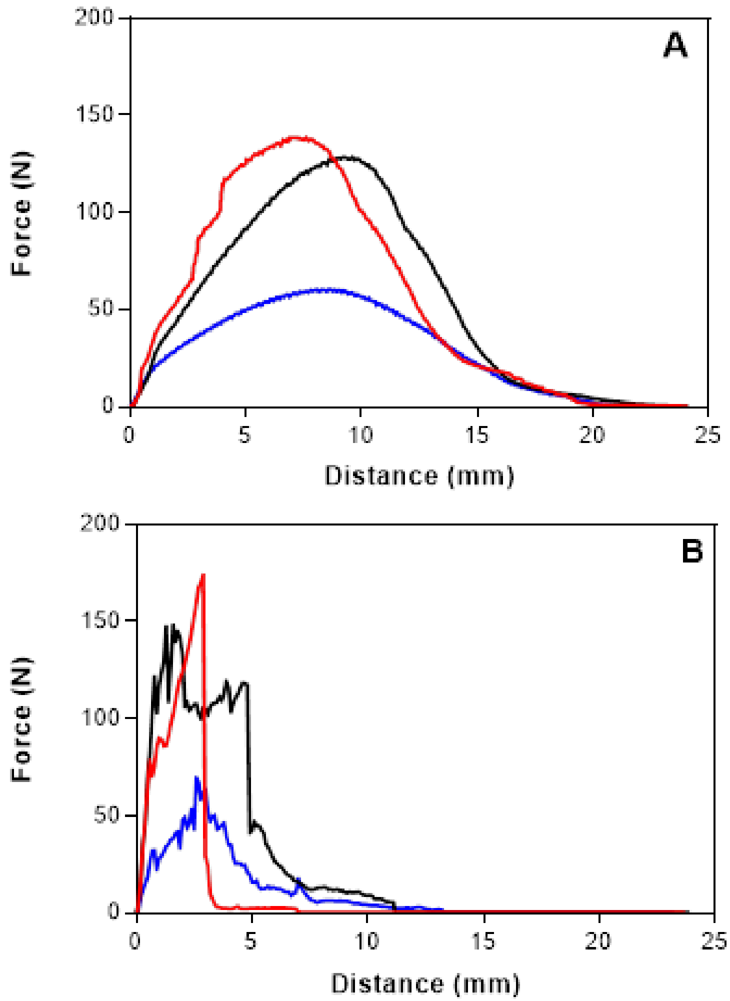

A
Force (N)
200
150
100
50
0
0
5
10
15
20
25
Distance (mm)
B
Force (N)
Distance (mm)

Figure 1

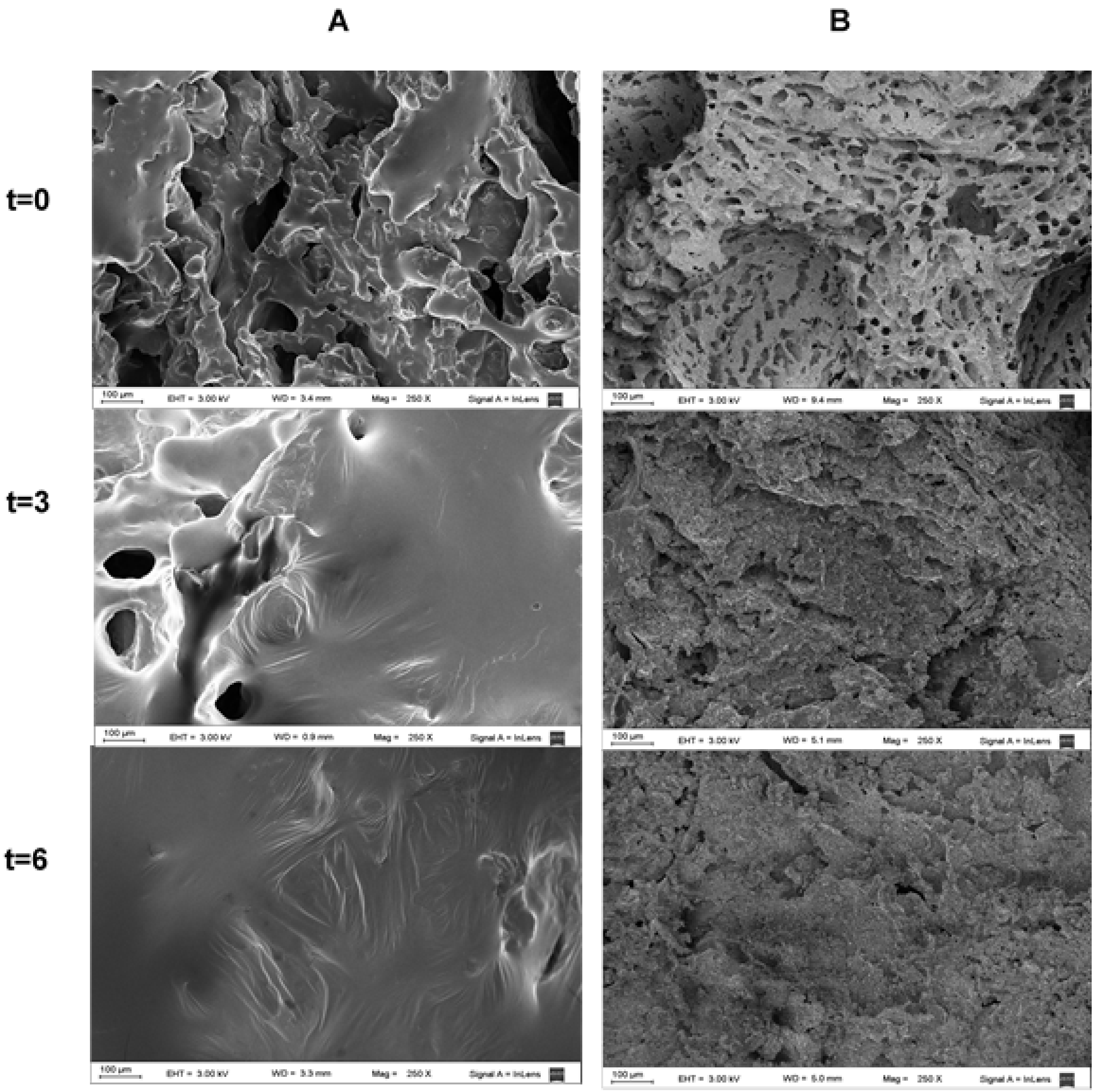
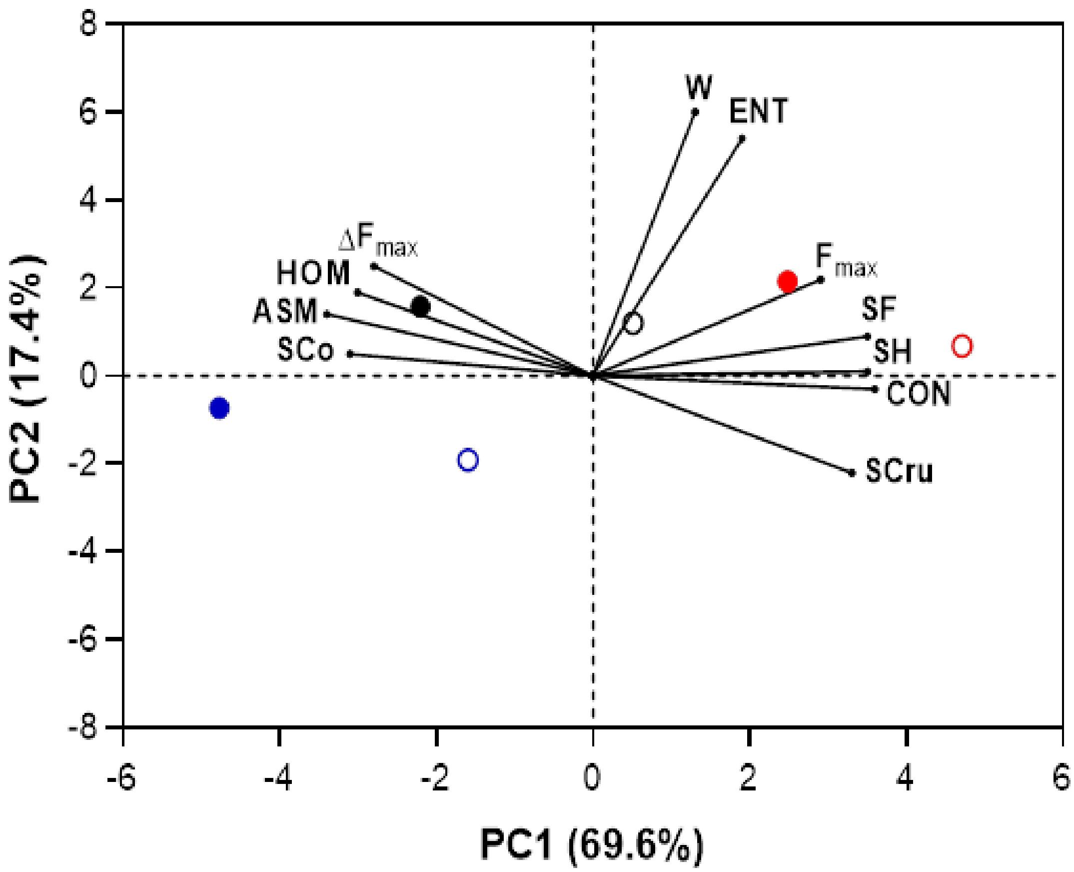


Figure 2



8
6
4
2
0
-2
-4
-6
-8
-6
-4
-2
0
2
4
6
PC2 (17.4%)
PC1 (69.6%)
W
ENT
ΔF_{max}
HOM
ASM
SCo
F_{max}
SF
SH
CON
SCru

Figure 3

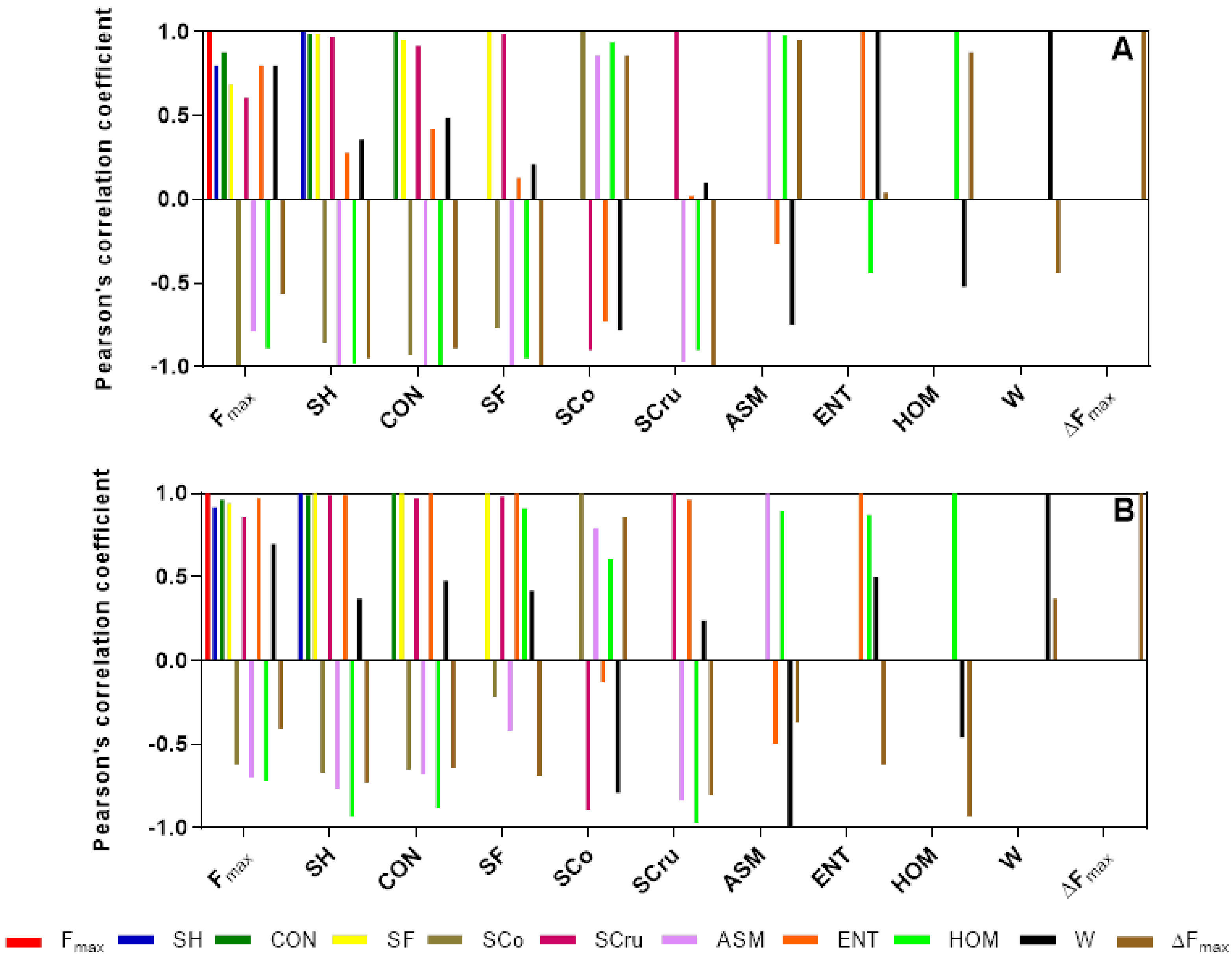

A
B
Pearson's correlation coefficient
1.0
0.5
0.0
-0.5
-1.0
F max
SH
CON
SF
SCo
SCru
ASM
ENT
HOM
W
ΔF max
Fmax SH CON SF SCo SCru ASM ENT HOM W ΔFmax

Figure 4

Table 1 - Reference scales for hardness, adhesiveness to palate, fracturability and cohesiveness (Hough & Contarini, 1994) and crunchiness (Farroni, 2011) used in the sensorial test during storage at 25ºC.

Textural Attributes	Reference scales
Hardness	Cream cheese (1), hard-boiled egg white (2.5), frankfurter (5), olive (6), processed cheese (7), peanut (9.5), chocolate (11), hard candy (17)
Adhesiveness	Margarine (1), peach jam (3), milk jam (6), spreading cheese (8), peanut butter (12)
Fracturability	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	Pudding (1), fruit candy (3), processed cheese (5), chewing candy (8), raisin (10), sugus® candy (12) chewing gum (15)
Crunchiness	Cereal bar (1), cracker (5), cornflake (10), honey and oat cheerios (12),thin toast bread (17)

Table 2: Textural parameters obtained for HI and IS candies through the instrumental test performed along storage at 25ºC.

	Time (months)	Textural parameters		
		F_{max} (N)	ΔF_{max} (mm)	W (J)
HI	0	139 ± 14^{a}	6.9 ± 0.6^{a}	1.00 ± 0.17^{a}
	3	129 ± 11^{a}	9.2 ± 1.1^{b}	1.18 ± 0.09^{a}
	6	61 ± 4^{b}	8.6 ± 1.3^{bc}	0.52 ± 0.02^{b}
IS	0	174 ± 16^{A}	2.8 ± 0.4^{A}	0.50 ± 0.03^{A}
	3	145 ± 7^{B}	1.8 ± 0.3^{B}	0.26 ± 0.02^{B}
	6	70 ± 6^{C}	3.4 ± 0.2^{AC}	0.17 ± 0.04^{C}

Different lowercase letters (HI) and uppercase letters (IS) indicate significant differences between means ($p < 0.05$).

Table 3: Textural changes perceived by the trained panel members for HI and IS candies during storage at 25°C for each attribute.

	Time (months)	Hardness		Fracturability		Cohesiveness		Crunchiness	
		Judges	Scale	Judges	Scale	Judges	Scale	Judges	Scale
HI	0	12.5	chocolate	4.41	cracker	3.39	fruit candy	5.93	cracker
	3	5.84	frankfurter	1.20	pudding	5.09	Processed cheese	0.24	cereal bar
	6	3.21	hard-boiled egg white	0.69	pudding	10.0	raisin	0.11	cereal bar
IS	0	14.6	hard candy	9.73	toast bread	2.00	fruit candy	12.2	honey and oat cheerios
	3	9.43	peanut	6.27	sweet biscuit	2.50	fruit candy	3.97	cracker
	6	6.31	olive	3.77	cracker	2.07	fruit candy	1.30	cereal bar

Table 4: Textural parameters obtained for HI and IS candies through the image test during storage at 25ºC.

	Time (months)	Textural parameters (GLCM)			
		CON	ENT	ASM	HOM
HI	0	0.45 ± 0.02[a]	4.71 ± 0.10[a]	0.30 ± 0.10[a]	0.78 ± 0.01[a]
	3	0.27 ± 0.08[b]	4.75± 0.10[a]	0.68 ± 0.06[b]	0.88 ± 0.04[b]
	6	0.14 ± 0.04[c]	4.7± 0.3[a]	0.83 ± 0.07[c]	0.96 ± 0.02[c]
IS	0	0.89 ± 0.10[A]	6.72 ± 0.12[A]	0.14 ± 0.02[A]	0.76 ± 0.02[A]
	3	0.76 ± 0.02[B]	6.64± 0.08[A]	0.13 ± 0.03[A]	0.73 ± 0.01[A]
	6	0.64 ± 0.09[C]	6.56± 0.08[A]	0.15 ± 0,01[A]	0.73 ± 0.02[A]

Different lowercase letters (HI) and uppercase letters (IS) indicate significant differences between means ($p < 0.05$).