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Research Article

Influence of Low Glycaemic Index Sweeteners on Antioxidant, Sensory, Mechanical, and Physicochemical Properties of a Watermelon Jelly

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The replacement of sucrose by new noncariogenic and low glycaemic index sweeteners (isomaltulose and tagatose) and the addition of natural watermelon juice in jelly have been assessed in terms of composition, texture, colour, antioxidant activity, microbiology, and sensory properties. These analyses were performed initially and after 15 days of storage. Furthermore, the values were compared with those obtained in the analyses of a commercial watermelon jelly. The results showed that the antioxidant activity increased with the storage time in the control sample and in samples combining isomaltulose and tagatose. In addition, noncariogenic and low glycaemic index sweeteners did not affect the instrumental texture. However, the colour changed, especially in the sample containing tagatose only. Finally, the dessert containing tagatose and isomaltulose in equal proportion achieved a similar score in the sensory evaluation as the commercial one, showing the feasibility of using these sweeteners to reformulate watermelon jelly.

1. Introduction

Watermelon (Citrullus lanatus) is a seasonal fruit whose surplus production is usually insufficiently exploited. Moreover, watermelon plays an effective role in reducing oxidative stress through phytochemical lycopene, is also high in other antioxidants, and has been linked to a decreased risk of coronary heart disease [1]. Currently, the public increasingly demands more low glycaemic index and noncariogenic products including fruits and vegetables in their formulations, along with new food-processing technologies for the manufacturing of such food products. One way of doing so is to add fruit to jelly. They are usually prepared with traditional sugars (sucrose, glucose, etc.). However, their consumption involves certain drawbacks for health (high caloric intake, increased glycaemic index, etc.). In this sense, the new guidelines of the World Health Organization establish a reduction in the consumption of simple sugars up to 5% of the total daily caloric intake for an adult with a normal body mass index. This is intended to reduce communicable diseases in children and adults, in particular weight gain, dental caries, and type two diabetes [2].

Nowadays, natural alternative sweeteners that are metabolized by the organism and have nutritional advantages can be found in the market, such as isomaltulose, tagatose, stevia. Isomaltulose is a natural sweetener present in honey and sugar cane juice and it was first commercialized as a food sweetener in 1980 [3]. Moreover, isomaltulose is a disaccharide for use as a carbohydrate source, which totally or partially replaces sucrose or other highly digestible carbohydrates. However, it is less glycaemic, less insulinemic, and noncariogenic [4]. Hydrogenated isomaltulose is known as isomalt or isomaltitol and this sugar alcohol has a very low glycemic index and is also noncariogenic but unlike isomaltulose has a reduced calorie value and an effect like dietary fibre in the gut [5]. Recently, several studies have been performed replacing sucrose by isomaltulose in sweet foods as gummies and marshmallows [6, 7]. Isomaltulose was recognized as safe (GRAS) in 2005 [8]. On the other hand, D-tagatose is a low carbohydrate functional sweetener, which is very similar in structure to fructose. Additionally, it is found naturally in cheese and yoghurt, and it can also be produced from D-galactose [9, 10]. However, it is metabolized differently, has a minimal effect on blood glucose and insulin levels,

and also provides a prebiotic effect [9, 11]. It can be used in ready-to-eat cereals, diet soft drinks, frozen yoghurt/nonfat ice cream, soft/hard confectionary, and marmalades [12-14]. The unabsorbed tagatose is fermented in the colon, where it acts as soluble fibre [15, 16]. Besides, it does not promote tooth decay and it only provides 1.5 kcal/g to diet [17]. Tagatose was generally recognized as safe (GRAS) in 2010 [18]. Consequently, the aim of this study was to assess the replacement of sucrose in jelly with low glycaemic and noncariogenic sweeteners (isomaltulose and tagatose) and the addition of fresh watermelon juice to their formulation. Composition, antioxidant activity, optical and mechanical properties, microbiological stability, and sensory evaluation were analyzed. Subsequently, the results were compared with a commercial jelly (containing sucrose, flavourings, and colorants).

2. Materials and Methods

2.1. Watermelon Jelly Formulations and Manufacturing Processes. The ingredients used to prepare the sample were watermelon juice (Citrullus vulgaris), mineral water (Aguas Danone S. A., Barcelona, Spain), sucrose (Azucarera Iberia S. L., Madrid, Spain), isomaltulose (Beneo-Palatinit, Mannheim, Germany), commercial tagatose (Tagatesse, Damhert NV/SA, Heusden-Holder, Belgium), and gelatine (Junca Gelatines, SL, Girona, Spain). Moreover, a commercial jelly in powder with watermelon flavourings (Royal, Kraft Foods, Madrid, Spain) was also characterized to compare the results. The following notation was used depending on the combination of sweeteners/sucrose used: control jelly: 100% sucrose; I50T50 jelly: 50% isomaltulose and 50% tagatose; T jelly: 100% tagatose; I jelly: 100% isomaltulose; and commercial jelly.

The recipe was prepared in accordance with the proportion of ingredients described in the commercial watermelon jelly box: 85% of sugars and 9.4% of gelatine. Moreover, the ingredients of commercial watermelon jelly were as follows: Vitamin C, flavourings and colorants (E100: curcumin and E120: carminic acid) and acidity regulators (fumaric acid, sodium citrate). It is noteworthy that in our new formulations of watermelon jelly no additives were used. Additionally, in the new formulations, 50% of mineral water was replaced with watermelon juice.

Watermelon was collected directly from crop. Later, in the laboratory, it was peeled and its juice was extracted (Molinex, model Vitapress, Mayenne, France). Then, the juice was mixed with the corresponding combination containing sucrose or a combination of sweeteners and water in a thermal blender (Thermomix, TM31, Vorwerk, Wuppertal, Germany) for 3.5 min at 100°C. Subsequently, the containers were filled and stored in a refrigerator at 4°C. All measurements were carried out in triplicate on the first day and after 15 days of storage.

2.2. Physicochemical Analyses. Soluble solid content (°Brix) was measured with a refractometer at 20°C (Atago 3T, Tokyo, Japan) and pH was registered with a pH meter (Seven Easy, Mettler Toledo, Barcelona, Spain), previously calibrated with

buffered solutions of pH 7.0 and 4.0. The moisture content $(x_w: g \text{ water/g watermelon jelly})$ was determined gravimetrically following an adaptation of the Official Methods of Analysis of AOAC International [19]. Water activity (a_w) was determined using a hygrometer (Decagon Devices, Inc., model 4TE, Pullman, Washington, USA).

- 2.3. Antioxidant Activity. The antioxidant activity of watermelon jelly was analyzed on the basis of the scavenging activities of the stable 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical following the protocol described in previous studies [20]. One gram of watermelon jelly was mixed with 6 ml of pure methanol for 5 min in a vortex, keeping the supernatant. This mixture was centrifuged at 13,000 rpm for 10 min. The absorbance was read at 515 nm in a spectrocolorimeter (Helios Zeta UV-VIS, Thermo Fisher Scientific, Inc., Waltham, MA, USA). Quantification was performed considering a standard curve of Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) and the results were expressed as mg of Trolox equivalent per 100 g of watermelon jelly.
- 2.4. Textural Characteristics of Watermelon Jelly. The instrumental texture measurements of the watermelon jelly were determined by TA.XT plus Texture Analyser (Stable Micro Systems, Godalming, UK), a Texture Profile Analysis Test (TPA). The test conditions involved two consecutive cycles of 50% compression with 15 s between cycles test speed of 1 mm/s. Moreover, a load cell of 50 kg and a 45 mm diameter cylindrical probe were used. Finally, the parameters of hardness, cohesiveness, adhesiveness, and springiness were obtained.
- 2.5. Optical Properties. The optical properties of watermelon jelly were measured using a spectrocolorimeter (Konica Minolta Inc., CM-3600d model, Tokyo, Japan) in 20 mm wide cuvettes. CIE- $L^*a^*b^*$ coordinates were obtained using D65 illuminant and 10° observer as the reference system. Lightness, a^* and b^* components, and Chroma (C^*) and hue (h^*) parameters were registered.
- 2.6. Microbiological Analysis. Yeasts and molds and mesophilic aerobics were determined. Serial dilutions were prepared by homogenizing 10 g of watermelon jelly with 90 mL of 1% sterile peptone water in a stomacher bag, using sterile techniques. Yeast and molds were determined in Sabouraud Chloramphenicol Agar (Scharlau Chemie, 1–166, Barcelona, Spain) plates kept for 5 days. Mesophilic aerobic populations were analyzed in a Plate Count Agar (Scharlau Chemie, 1–329, Barcelona, Spain), by incubating samples for 72 h at 31°C. Microbial counts were expressed as CFU/g. Plates were inoculated in triplicate. Samples were taken for analyses on days 1 and 15.
- 2.7. Sensory Evaluation. Consumer preferences were evaluated using an acceptance test 9-point hedonic scale according to the methods identified by the International Standards Organization [21], the following attributes in the samples: colour, flavour, texture, sweetness, intention of buying, and

Table 1: Values for moisture content (x_w : g water/g watermelon jelly), "Brix, water activity (a_w), pH, and antioxidant activity (mg Trolox/100 g watermelon jelly) of watermelon jelly formulated with sucrose (control and commercial) or with sweeteners (isomaltulose and tagatose), initially and after 15 days of storage.

Formulation	Time (days)	x_w (g water/g watermelon jelly)	°Brix	a_w	рН	Antioxidant activity (mg Trolox/100 g watermelon jelly)
Commercial	1	0.852 ± 0.001^{a}	16 ± 0.4^{d}	0.996 ± 0.001^{c}	3.667 ± 0.006^a	65 ± 9 ^d
	15	0.850 ± 0.001^{a}	15.7 ± 0.2^{d}	0.994 ± 0.001^{b}	3.80 ± 0.02^{b}	$55 \pm 5^{\mathrm{e}}$
Control	1	0.853 ± 0.002^{a}	16.27 ± 0.06^{cd}	0.994 ± 0.0002^{b}	$6.337 \pm 0.006^{\rm e}$	7.9 ± 1.2^{ab}
	15	0.849 ± 0.003^{a}	16.27 ± 0.15^{c}	0.991 ± 0.001^{a}	$6.46 \pm 0.02^{\rm f}$	16.2 ± 0.2^{c}
I	1	0.863 ± 0.001^{b}	14.4 ± 0.2^{a}	0.993 ± 0.001^{ab}	6.203 ± 0.006^{d}	8.8 ± 1.3^{b}
	15	0.864 ± 0.005^{b}	14.73 ± 0.06^{ab}	0.993 ± 0.001^{ab}	$6.37 \pm 0.04^{\rm f}$	2.4 ± 1.1^{a}
T	1	0.860 ± 0.002^{b}	15.03 ± 0.15^{b}	0.992 ± 0.001^{ab}	6.25 ± 0.02^{d}	8.3 ± 0.7^{b}
	15	0.865 ± 0.003^{b}	15.1 ± 0.1^{b}	0.993 ± 0.002^{ab}	$6.40 \pm 0.03^{\rm f}$	3.7 ± 1.5^{ab}
I50T50	1	0.863 ± 0.001^{b}	14.8 ± 0.2^{b}	0.993 ± 0.0005^{b}	6.113 ± 0.013^{c}	9.9 ± 0.5^{b}
	15	0.873 ± 0.006^{c}	14.77 ± 0.21^{b}	0.994 ± 0.002^{b}	6.27 ± 0.04^{e}	15.8 ± 1.2^{c}

Equal letters indicate homogeneous groups.

global preference [22]. The panel consisted of 30 trained panellists in a 20–50 age group consisting of regular consumers of this kind of dessert. Testing sessions were conducted in a sensory evaluation laboratory built according to the international standards for test rooms [23]. In this study, the watermelon jelly elaborated only with isomaltulose (I) was not considered in the sensorial analyses since the other samples of jelly showed better quality to determine the consumers' preference.

2.8. Statistical Analysis. Multifactor ANOVAs were performed using a multiple comparison test and a LSD test ($\alpha = 95\%$), with Statgraphics Centurion software (Statpoint Technologies, Inc. Warrenton, Virginia, USA). Interactions between the factors were also considered.

3. Results and Discussion

3.1. Physicochemical Analyses. The moisture content results (x_w) , Brix, pH, water activity (a_w) , and the antioxidant activity of the jelly formulations with sucrose (control and commercial) or low glycaemic index sweeteners (tagatose and isomaltulose) are shown in Table 1. In all cases the jelly reached a concentration of soluble solids higher than 14° Brix, being higher in the commercial and control samples. Consequently, the samples made with sweeteners showed a higher content of water. Despite the significant statistic differences found in terms of water activity among the various jelly, in all cases the a_w values were 0.99. Therefore, the type of sweetener had no influence on this parameter. However, the pH was much lower in the commercial jelly due to the acidity regulators (fumaric acid and sodium citrate) used in its formulation. Time did not cause significant changes in any of these parameters, except for the pH, which slightly increased after 15 days of storage. These results are coherent with those established in other studies on gummy confections [7, 24]. Regarding the antioxidant activity, initially the commercial sample had the highest value due to the presence

of vitamin C. On the other hand, the samples prepared with watermelon juice initially showed the same antioxidant activity but increased in the control and I50T50 samples over time. Thus, the mixture of isomaltulose and tagatose could have a synergetic effect favouring antioxidant concentration. Liu [25] observed that the reactions which occur between antioxidant compounds may be synergistic or additive, which is why the measurement of antioxidant activity could offer a global estimation of contribution of the different compounds to global antioxidant activity. Moreover, we observed a significant reduction of antioxidant activity during the storage period in the rest of the samples studied. Rababah et al. [26] obtained similar values in orange marmalade due to the oxidation of the components responsible for this activity. Besides, from a theoretical estimation, according to the values of glycaemic (GI) of fructose (20), sucrose (60), isomaltulose (32), and tagatose (0), the theoretical GI of the watermelon jelly formulations has been calculated. These estimations have been obtained by applying the following mathematical relation: $GI^{theoretical} = \sum m_i \cdot GI_i / = \sum m_i$, with m_i being grams of each component and GIi being the glycaemic index of each component. Thus, values of GI^{theoretical} of the commercial formulation of watermelon jelly would be 7.9, in the case of jelly prepared only with sucrose in terms of sugars, but, with watermelon juice, this index would be 5.7, when the only sugar would be isomaltulose, GI^{theoretical} = 3.1, for only tagatose, $GI^{theoretical} = 0.3$, and when the amount of sugars was in the same proportion for tagatose and isomaltulose, GI^{theoretical} = 1.7. In this regard, the percentage of GI reduction for the jelly with watermelon juice and sucrose would be around 28%, when isomaltulose was used around 60%, for tagatose 96%, and finally for combination of isomaltulose and tagatose in the same amount, around 78%. In any case, these values should be checked with medical tests.

3.2. Textural Characteristics of Watermelon Jelly. Figure 1 shows the average curves of the TPA analysis carried out

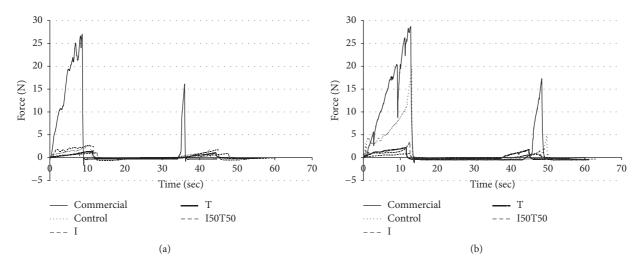


FIGURE 1: Representative curves of TPA test for watermelon jelly studied as a function of sweeteners used in its formulation, initially (a) and after 15 days of storage (b).

on the samples of jelly used in this study, along with the graphics used to determine the mechanical parameters. As can be observed, the curves obtained for the commercial jelly showed more pronounced peaks than those of other samples, both at the beginning and at the end of the storage period. Furthermore, after 15 days of storage, the second peak of the curve shifted to the right, especially in the commercial jelly. The interaction charts ($\alpha = 95\%$) of the mechanical parameters are shown in Figure 2. Initially, there were no significant differences in any mechanical parameters between the different samples of jelly, except for the springiness of the commercial jelly, which was significantly lower. However, after 15 days of storage, the commercial jelly showed a significant increase in hardness and gumminess, while the control samples showed a decrease in adhesiveness, cohesiveness, and springiness. The jelly formulated with new sweeteners maintained all its mechanical properties over time. Moreover, the best score was achieved by the mixture of isomaltulose and tagatose. However, other authors [6], in studies carried out in gummy confections, recorded lower values of hardness when isomaltulose was used as compared to samples prepared with sucrose and glucose syrup before being stored. Therefore, according to these results, the low glycaemic index sweeteners used in this study could have a greater ability to maintain the mechanical properties over time.

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3.3. Optical Properties. Figure 3 shows the interaction charts of the colorimetric coordinates L^* , a^* , and b^* , Chroma (C^*) and hue (h^*) as a function of the formulation used and the storage time. As can be seen, the commercial jelly showed significant differences in coordinates L^* , a^* , and b^* as compared to the jelly samples formulated with low glycaemic index sweeteners. Concretely, commercial samples showed lower values of luminosity but higher values of a^* and b^* coordinates. These differences may be attributable to the colorants used in commercial jelly (carminic acid (E-120) and curcumin (E-100)). Considering only the jelly formulated with watermelon juice, it was noteworthy that the

use of the new sweeteners significantly reduced its luminosity. Besides, in the formulation containing only tagatose, L^* significantly decreased over time. Regarding the a* coordinate, initially there were no significant differences among the samples. However, after the storage period, the sample of jelly containing only tagatose presented a significant change once again, but, in this case, an increase. The b^* coordinate followed the same trend as the a^* coordinate. In this case, the mixture of isomaltulose and tagatose also showed an increase after storage. As a consequence, Chroma (C^*) was significantly higher after 15 days in samples that only contained tagatose. Finally, the major values of hue (h^*) were recorded in the I50T50 jelly. As reported by Peinado et al. [27], in general, storage induced a reduction of L^* , a^* , and b^* parameters in spreadable strawberry products. Consequently, lower Chroma (C^*) and a slight decrease in hue (h^*) were observed too.

3.4. Microbiological Analyses. Initially, microbial counts of mesophilic aerobics, yeasts, and molds were not found in samples. However, at the end of storage, mesophilic aerobics, yeasts, and molds were found in new formulations. Nevertheless, the microbial counts must not exceed $5 \cdot 10^1$ CFU/g yeasts and molds and $5 \cdot 10^2$ CFU/g mesophilic aerobics [28]. In summary, the new formulations of watermelon jelly were microbiologically stable during the storage period. It is noteworthy that we did not use additives, in contrast with the commercial sample, which used acidity regulators (fumaric acid and sodium citrate) in its formulation.

3.5. Sensory Analyses. The results of the sensory analyses of jelly for the different formulations (commercial, control, T, and I50T50) are presented in Figure 4. In this case, two ANOVAs were performed in order to assess the influence of the commercial jelly on these results. Thus, one ANOVA was carried out considering all formulations (commercial, control, T, and T50I50) and the other without considering the commercial sample (control, T, and T50I50). The results obtained in the first ANOVA revealed a better assessment of

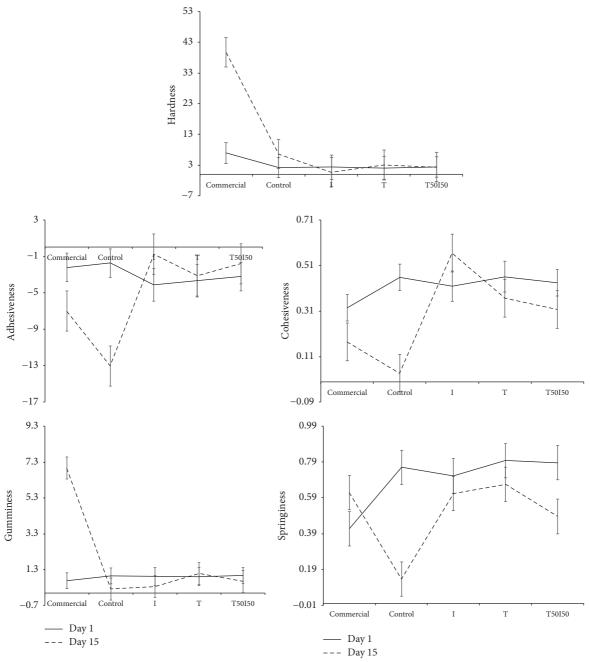


FIGURE 2: Interaction graphics ($\alpha = 95\%$) of hardness, adhesiveness, cohesiveness, gumminess, and springiness of watermelon jelly as a function of the formulation and storage time.

the colour and flavour in commercial jelly in comparison to the other samples of jelly due to the addition of colorants and artificial flavours, as commented earlier. Moreover, the texture of the jelly made with a mixture of isomaltulose and tagatose presented values similar to the commercial jelly, being significantly lower in the other two cases. Finally, no significant differences were observed with respect to sweet taste, global acceptance, and intention of buying between the samples of jelly studied in the ANOVAs analyzed. These results are coherent with those established in other studies on gummy confections [7]. The total sugar content of marshmallows could be replaced by a mixture of isomaltulose and

fructose in equal proportions. These marshmallows obtained a better sensory evaluation than those confected with sucrose and glucose syrup.

4. Conclusions

The reformulation of watermelon jelly with low glycaemic index sweeteners used in this research is viable. Regarding antioxidant activity, the mixture of isomaltulose and tagatose enhanced its levels during storage, achieving similar values to those of the jelly containing sucrose, though not reaching the values of commercial jelly. Moreover, the mechanical

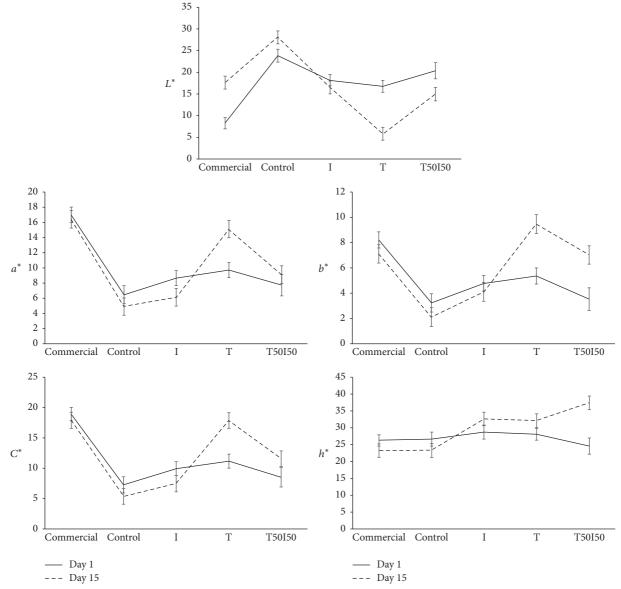


FIGURE 3: Interaction graphics ($\alpha = 95\%$) of colour parameters: L^* , a^* , and b^* coordinates, Chroma (C^*) and hue (h^*) of the watermelon jelly as a function of the formulation and storage time.

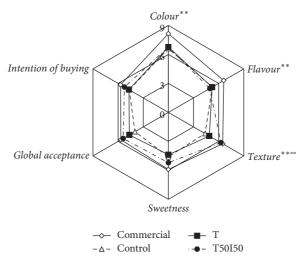


FIGURE 4: Results of the sensory analysis of watermelon jelly as a function of the formulation. Considering all jelly: **P value < 0.01 and without the commercial jelly: **P value < 0.01.

properties of the jelly made with noncariogenic and low glycaemic index sweeteners were similar to those of the jelly prepared with sucrose and remained constant. On the other hand, further studies should be carried out in order to improve over time the colour stability of the jelly formulated with tagatose over time. Nevertheless, the watermelon jelly formulated with new sweeteners was microbiologically stable. In terms of sensory analyses, texture was a determinant attribute in the evaluation of the watermelon jelly. Finally, the jelly containing equal proportions of isomaltulose and tagatose and the commercial jelly achieved the best scores.

Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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