PRETREATMENT BEHAVIOR OF FROZEN STRAWBERRIES AND STRAWBERRY PUREES FOR SMOOTHIE PRODUCTION

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Abstract

Recently, for increasing fruits shelf life, food industries used thermal processing, which has been shown in some studies to affect the sensorial and physic-chemical properties of these products. During smoothie production is mandatory to have raw materials for the whole year, so it is necessary to storage it after different pretreatments such as freezing. Color, flavor, texture and physic-chemical properties of food products have an important role in correlation with taste, sensory perception and consumer acceptance. These are critical quality attributes affecting the acceptability of fruits, fresh, frozen or processed (concentrates, jam, juice, nectar, syrup, dairy products), thus being of major concern in product design.

Strawberries are popular fruit with high visual appeal and desirable flavor, but are highly perishable, being susceptible to mechanical injury, water loss, decay and physiological deterioration. The natural color of strawberries and their products easily fades or deteriorates during processing and storage.

The aim of this study is to observe the influence of freezing storage on color, texture, antioxidant activity, vitamin C and physic-chemical properties of strawberry and strawberries puree.

For this purpose, were realized and analysed 4 different samples starting with just harvested strawberries, pressed strawberries, strawberries immersed in ascorbic acid 1% for 5 minutes and strawberries blanched at 95°C for 5 minutes, before and after freezing. These purees were stored for 6 months in the freezer. During frozen storage the lightness index, L, yellowness index, b and the redness index, a, were also measured and it was observed colour changes for all of strawberry samples. pH and acidity values, showed that the strawberry samples registered insignificant changes compared to standard (control) samples.

Key words: color, strawberry, freezing, shelf life.

INTRODUCTION

Previous studies have shown that the fruits and vegetables contain high levels of antioxidant compounds that have proven capacity of reducing oxidative stress responsible for DNA, protein and membrane damage (Mandavea et al., 2014). It therefore lower the incidence and mortality rates of various cancers and heart diseases (Cao et al., 1996; Bagchi and Puri, 1998; Lobo et al., 2010; Sen and Chakraborty, 2011). Strawberry is one of the most popular summer fruit worldwide that are characterized with unique and highly desirable taste and flavour (organic acids and soluble sugars content), color (Campaniello, Bevilaqua, Sinigaglia, & Corbo, 2008; Koyuncu & Dilmaçünlü, 2010) and texture (Velickova et al., 2013). Strawberries are a good source of vitamin C (Patras, Brunton, Da Pieve, & Butler, 2009), being available worldwide as a whole fruit and also as a puree/juice/concentrate to be incorporated in nectar, ice cream, yoghurt, baby food and confectionary (Sulaiman and Silva, 2013). Is a widely researched fruit for nutritional and health benefits and organoleptic properties. This fruit is rich in vitamins, minerals, fibre and phytochemicals. In addition, strawberries contain potentially bioactive compounds and are a great source of phenolic compounds such as flavonoids and phenolic acids (Aaby, Skrede, & Wrolstad, 2005; Määttä-Riihinen, Kamal-Eldin, & Törnönen, 2004; Seeram, Lee,
Scheuller, & Heber, 2006). These compounds make strawberries a highly antioxidant fruit (Aaby et al., 2005; Wolfe et al., 2008) with potential health benefits. Among the numerous healthy properties described in the literature are anti-proliferative effects on cancer cells (Meyers, Watkins, Pritts, & Liu, 2003; Olsson, Andersson, Oredsson, Berglund, & Gustavsson, 2006) and the antioxidant and anti-inflammatory effects that have been shown to reduce cardiovascular disease risk factors in several prospective cohort studies (Hannum, 2004). Also, strawberries may exert protection against inflammation, type 2 diabetes, cardiovascular disease, hypertension, oesophageal cancer, obesity in humans (Cassidy et al., 2011; Chen et al., 2012; Giampieri et al., 2012) and oxidative stress (Kanter et al., 2012). Cheel et al. (2007) demonstrated that strawberry extracts had significantly higher antioxidant activity compared to 11 other fruits. Qualitative and quantitative variations in the antioxidant activity have been observed among strawberry cultivars as well as within the same variety, depending on the genetic background, degree of ripening, postharvest storage of the fruits, and climatic factors (Maatta et al., 2004; Lopes-da-Silva et al., 2007). However strawberries are very problematic for industrial processing as they are seasonal, and have a high water content which makes them very perishable (Oey et al., 2008; Peinado et al., 2013; Taiwo et al., 2003). Furthermore, color plays a major role in quality assessment of food significantly determining consumers’ choice (Stintzing & Carle, 2004). Accordingly, strawberries are commonly frozen, thus allowing a year-round production of jams, juices, fruit preparations, purées, concentrates (Oszmiański, Wojdylo, & Kolińak, 2009; Skrede, 1996) and smoothies. Although freezing is an efficient preservation measure (Holzwarth et al., 2012, Singh & Wang, 1977), it is inevitably accompanied by irreversible structural damage of the cell wall, middle lamella, and protoplast, resulting in textural quality losses (Van Buggenhout, Sila, Duvetter, Van Loey, & Hendrick, 2009). Therefore, the aim of this study is to observe the influence of freezing storage on color, texture, antioxidant activity, vitamin C and physio-chemical properties of strawberry and strawberries puree used for smoothie production.

MATERIALS AND METHODS

Samples
Strawberries were purchased from a local market from Romania, Bucharest. All fruits were selected based on the same ripening stage (>90% red surface color), uniform size, absence of any physical damage and fungal infection. They were either processed immediately and packed for freezing within 2 h, or analyzed. Strawberries were packed in 200 g plastic bags and stored for 6 months at -20°C. Were realized 4 different samples starting with just harvested strawberries, pressed strawberries, strawberries immersed in ascorbic acid 1% for 5 minutes and strawberries blanched at 95°C for 5 minutes. These were analysed before and after freezing. Frozen strawberries were kept for 24 h at 4°C before they were analyzed.

Physico-chemical analysis

pH determination
pH was determined with a pH meter WTW INOLAB 720 series type with automatic temperature compensator, whose pH domain is between 0,00-14.00, with a precision of ± 0,01.

Titratable acidity (TA)
Titratable acidity was determined by titrating 10 g of homogenized sample with 0.1 N NaOH to an end point of pH 7.3 using Schott automatic titrator type Titronic basic. TA was analyzed in duplicates and expressed as citric acid/100 g product (factor 0.64).

Brix degree, dry matter content (S.U. %) and water activity (a_w) determination
The level of sugars was measured as Brix by a Krüss Refractometer and correlated with the amount of soluble solids (expressed as sucrose concentration) using the conversion table or read directly on the scale Refractometer. The
dry matter content was determined after drying approximately 5 g of pulp at 140°C till a stable weight, with Precisa XM 60 thermobalance. Water activity was conducted with special system Novasina.

**Color**

Color assessment of the samples was conducted at room temperature using a HunterLab colorimeter, Miniscan XE Plus. This instrument was calibrated using the black and white tiles provided. Instrumental color was measured using Illuminant D65 and 10° observer angle. Samples were filled into a low reflectance sample container and placed over the colorimeter chamber. For each sample, measurements were made in ten different points and results were averaged. Therefore the total color change (ΔE) was calculated with the following equation (Hunter Lab, 1996):

$$\Delta E = [(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2]^{1/2}.$$ 

**Antioxidant activity**

The effect of antioxidant activity on DPPH was estimated according to the procedure described by Villaño, Fernández-Pachón, Moyá, Troncoso, & García-Parrilla, 2007, with some modification. To obtain DPPH solution (60 µM), 2.36 mg DPPH were diluted in 100 ml ethanol. Samples were diluted appropriately in ethanol. Sample preparation was done by maceration in ethanol (75%) for 2-3 days in the dark at room temperature. All measurements were performed in triplicate. For each measurement, 0.05 ml sample in ethanol was added to 1.95 ml DPPH ethanolic solution (60 µM). These solutions were vortexed thoroughly, and incubated in dark at room temperature for 30 min (Gülçin, 2010). After 30 min, sample absorbance was measured at 515 nm (t=30min) against DPPH ethanolic solution alone (t=0 min). For calibration curve were used six different concentrations of Quercitin (100-3.125 µM). Absorbance measurements were recorded on a UV/Vis spectrophotometer Unicam Helios Gamma. Results were expressed as quercitin equivalents using the following equations:

$$\text{AA}_R \text{ (QE)} = \text{A}_R \text{ (Quercitin equivalents)}$$

%ΔA<sub>515</sub> = [(A<sub>515</sub> (t=0)-A<sub>515</sub> (t=30))/A<sub>515</sub> (t=0)] x 100

**Ascorbic acid spectrophotometric determination**

The content of ascorbic acid from strawberry samples was determined by using a UV/VIS spectrophotometer Unicam Helios Gamma at 500 nm. 10 g of fruit pulp was extracted with 30 ml of 1% oxalic acid in a homogenizer for 1 min. The extract was filtered through a filter paper. After filtration, 2 ml from extract solution, 1 ml oxalic acid 1%, 5 ml tampon solution, 2 ml indophenol (2, 6-Dichlorophenol Indophenol) and 20 ml xylene, were placed in a centrifuge tube and centrifuged 20 min at 4°C and 9000 rpm. After absorbance measurements, ascorbic acid content was expressed in milligrams at 100 g product, and is calculated with following equation:

$$\text{Vit.C(mg/100g)} = \frac{(V_0 - V_1) \times V_3 x C/(V_4 \times V_2) x 100}{V_0 - V_1}$$

where:
- $V_0$ - indophenol solution volume added for reduction,
- $V_1$ - indophenol solution excess volume read on the standard curve,
- $V_2$ - sample volume for analysis,
- $V_3$ - sample volume brought for analysis,
- $V_4$ – acid extract volume used for analysis,
- $C$ - ascorbic acid corresponding quantity for 1 ml indophenol solution.

**RESULTS AND DISCUSSIONS**

**Evolution of physical chemical properties**

The obtained results showed that the pH value of the samples registred insignificantly changes. The highest difference was recorded at just harvest strawberry samples (fresh: 3.6 (±0.01) and frozen: 3.9 (±0.02)). In this case, pH value is apparently not responsible for color changes during on freezing storage as it also observed by Gössinger et all. (2009). Acidity, expressed as citric acid content, which is the main acid of strawberries (Kamperidou & Vasilakakis, 2006), ranged between 0.59 (±0.1) (P1 before freezing) and 0.9 (±0.01) (P2 after freezing). Samples of frozen stored
strawberry had higher acidity than those fresh, and is high correlated with Brix values, which decrease (Galoburda et al., 2014) after freezing storage for 6 months (Table 1, Figure 2 and 3). It means that acidity and Brix are responsible for color changes during on freezing storage. For dry matter content, three strawberry samples (P2, P3 and P4) recorded significant decreases from 56.07% to 18.48% for blanched strawberries before and after freezing (P4) (Table 1, Figure 4). Trough processing method, first samples (P1 – just harvested strawberry) recorded insignificant changes for dry S.U. and aw (Table 1, Figure 4 and 5).

Table 1. Physico-chemical results of strawberries before and after freezing (P1 – just harvested strawberries, P2 – pressed strawberries, P3 – strawberries immersed in ascorbic acid 1% for 5 minutes, P4 – strawberries blanched at 95°C for 5 minutes)

<table>
<thead>
<tr>
<th>Sample</th>
<th>pH Before</th>
<th>pH After</th>
<th>Titratable acidity (g citric acid/100g product) Before</th>
<th>Titratable acidity (g citric acid/100g product) After</th>
<th>Brix Before</th>
<th>Brix After</th>
<th>aw Before</th>
<th>aw After</th>
<th>S.U. % Before</th>
<th>S.U. % After</th>
</tr>
</thead>
<tbody>
<tr>
<td>P1</td>
<td>3.6</td>
<td>3.9</td>
<td>0.59</td>
<td>0.74</td>
<td>11.1</td>
<td>10.3</td>
<td>0.932</td>
<td>0.928</td>
<td>69.33</td>
<td>60.93</td>
</tr>
<tr>
<td>P2</td>
<td>3.5</td>
<td>3.6</td>
<td>0.81</td>
<td>0.9</td>
<td>9.8</td>
<td>9.2</td>
<td>0.932</td>
<td>0.931</td>
<td>53.36</td>
<td>18.52</td>
</tr>
<tr>
<td>P3</td>
<td>3.4</td>
<td>3.5</td>
<td>0.75</td>
<td>0.83</td>
<td>7.8</td>
<td>7.4</td>
<td>0.932</td>
<td>0.923</td>
<td>52.59</td>
<td>18.46</td>
</tr>
<tr>
<td>P4</td>
<td>3.6</td>
<td>3.7</td>
<td>0.63</td>
<td>0.64</td>
<td>8.5</td>
<td>8.3</td>
<td>0.948</td>
<td>0.933</td>
<td>56.07</td>
<td>18.48</td>
</tr>
</tbody>
</table>

Figure 1. Variation of the strawberries pH values before and after freezing (for 6 months)

Figure 2. Variation of the strawberries Brix degree values before and after freezing (for 6 months)

Figure 3. Variation of the strawberries titratable acidity values before and after freezing (for 6 months)

Figure 4. Variation of the strawberries dry matter content values before and after freezing (for 6 months)
Besides texture and economic considerations, color is one of the most important factors in the perception of strawberry fruit quality, affecting consumer acceptance (Abd-Elhady, 2014). Color stability of strawberry products, particularly after heat or cold and light exposure, remains a challenge (Carle et al., 2001). In general, several factors are believed to affect the color and stability of strawberry anthocyanins include structure and concentration, pH, temperature, light, presence of co-pigments, self-association, metallic ions, enzymes, oxygen, ascorbic acid, sugar and their degradation products, proteins, and sulfur dioxide (Rhim, 2002). During the 6 months of freezing storage period, no visually detectable color changes were observed for any of the analyzed strawberry samples. Changes were observed when the color characteristics were analyzed with colorimeter Hunter Lab according to Universal Software V4.01 MiniScan™ XE Plus program. During freezing storage period, the L (lightness), a (redness) and b (yellowness) values of just harvested strawberries (P1), pressed strawberries (P2), strawberries immersed in ascorbic acid 1% for 5 minutes (P3) and strawberries blanched at 95°C for 5 minutes (P4) tended to decrease, indicating color changes, as can be observed in figures 6, 7, 8 and 9. For all strawberry samples (P1, P2, P3, and P4), the L (lightness), a (redness) and b (yellowness) values tended to decrease during freezing period, indicating a discoloration of the samples (figures 6, 7, 8 and 9). This are in correlation with increasing acidity and decreasing Brix for freezing storage period.

The ΔE values, which are an indicator of total color difference (table 2), showed that freezing storage for 6 months affected color attributes for all strawberry samples.

### Table 2. Instrumental color variables of strawberry samples before and after freezing

<table>
<thead>
<tr>
<th></th>
<th>L</th>
<th>a</th>
<th>b</th>
<th>ΔE</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before</td>
<td>After</td>
<td>Before</td>
<td>After</td>
</tr>
<tr>
<td>P1</td>
<td>26.7</td>
<td>24</td>
<td>30.8</td>
<td>28.65</td>
</tr>
<tr>
<td>P2</td>
<td>21.83</td>
<td>19.84</td>
<td>29.83</td>
<td>24.96</td>
</tr>
<tr>
<td>P3</td>
<td>22.67</td>
<td>20.85</td>
<td>30.62</td>
<td>25.65</td>
</tr>
<tr>
<td>P4</td>
<td>31.62</td>
<td>31.55</td>
<td>28.11</td>
<td>25.91</td>
</tr>
</tbody>
</table>

Figure 6. Graphical representation of the values of L, a, b, according to Universal Software V4.01 MiniScan™ XE Plus program for just harvested strawberry samples before and after frozen 6 months.
Antioxidant activity

DPPH assay is one of the best known, frequently employed and accurate method of assessing free radical scavenging activity. DPPH is a stable free radical because of its spare electron delocalization over the whole molecule. Decolourization causes deep violet color with maximum around 520 nm. When a solution of DPPH is mixed with a substrate acting as a hydrogen ion donor, a stable non-radical form of DPPH is obtained with simultaneous change from violet to pale yellow (Mandave et al., 2014). Antioxidant activity content in just harvest strawberry samples (P1)
decreased from 1069.26 μM quercitin equivalents to 1045.08 μM quercitin equivalents for frozen sample (P1). Highest value for DPPH content it was seen at fresh strawberries immersed in ascorbic acid 1% for 5 minutes (1128.31 μM quercitin equivalents) but also here it was recorded the most significant decrease like 1011.96 μM quercitin equivalents. For blanched strawberry samples, DPPH values recorded insignificant changes like from 1085.48 μM quercitin equivalents to 1082.74 μM quercitin equivalents after freezing sample. In conclusion, antioxidant capacity of blanched strawberry samples (95°C fir 5 min) is more stable (fresh: 1085.48 μM quercitin equivalents and frozen: 1082.74 μM quercitin equivalents) than samples immersed in ascorbic acid 1% for 5 minutes (fresh: 1128.31 μM quercitin equivalents and frozen: 1011.96 μM quercitin equivalents).

![Figure 10. Variation of antioxidant activity for strawberry samples before and after freezing storage](image)

Vitamin C (Ascorbic Acid spectrophotometric determination)
Generally, fruits and vegetables show a gradual decrease in vitamin C content as the storage temperature or duration increases (Koyuncu & Dilmaçünal, 2010). Vitamin C content in just harvest strawberry samples (P1) decreased from 26.21 (±0.02) mg Vit C/100 g product up to 26.06 (±0.02) mg Vit C/100 g product for frozen sample (P1). Lowest values for vitamin C content was recorded from pressed strawberry samples (P2) from 20.62 (±0.07) mg Vit C/100 g product to 20.19 (±0.03) mg Vit C/100 g product. All changes in vitamin C content of strawberries during freezing storage is shown in Figure 11. Similar results were found by Koyuncu & Dilmaçünal, 2010, and Galoburda, 2014. As a result strawberry purees contains smaller amount of vitamin C compared to whole strawberry fruits.

![Figure 11. Variation of vitamin C content of strawberry samples before and after freezing storage](image)
CONCLUSIONS

During smoothie production is mandatory to have raw materials for the whole year, so it is necessary to storage it after different pre-treatments such as freezing.

Color is one of the most important factor in the perception of strawberry fruit quality, affecting consumer acceptance and preference, but only after texture and economic considerations.

During the 6 months of freezing storage period were not observed visually detectable color changes for any of the analysed strawberry samples. No significant changes were observed when the color characteristics were analysed with colorimeter.

The obtained results showed that the pH value of the strawberry samples registered insignificantly changes, during the freezing period.

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REFERENCES


Kamperidou I., M. Vasilakakis M., 2006, Effect of propagation material on some quality attributes of strawberry fruit (Fragaria x ananassa, var. Selva), Scientia Horticulturae 107, 137–142.


