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Ultrasound assisted osmotic dehydration of organic cranberries (Vaccinium oxycoccus): Study on quality parameters evolution during storage

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Ultrasound assisted osmotic dehydration of organic cranberries (*Vaccinium oxycoccus*): Study on quality parameters evolution during storage.

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- Title: Ultrasound assisted osmotic dehydration of organic cranberries (*Vaccinium oxycoccus*):
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- 3
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- 18

#### 19 Abstract

Cranberries are appreciated for their high amount of antioxidants such as flavonoids, anthocyanins, 20 phenolic acids, carotenoids and vitamins. However, due to their sour and tart taste they request to be 21 processed into sweeter dried fruits in order to be acceptable for the consumers. The aim of this work 22 was to analyse the effect of ultrasound assisted osmotic dehydration on mass transfer parameters and 23 on quality characteristics during storage of cranberries. Ultrasound treatment was performed at the 24 frequency of 21 kHz for 30 min in three osmotic solutions - 61.5% sucrose, 30% sucrose with an 25 addition of 0.1% of steviol glycosides and 40 % trehalose on cut in half cranberries. Afterwards, the 26 cranberry samples were subjected to osmotic dehydration process at 40°C for 72 h. The 27 28 osmodehydrated samples both with or without ultrasound pre-treatment were collected and stored at 10°C in climatic chamber in microperforated plastic bags (PLA) for 8 weeks. The weight reduction, 29 dry matter, water activity, colour, and microbial analysis were performed after 1, 2, 4 and 8 weeks of 30 31 storage. The obtained results indicated that ultrasound application significantly affected the mass transfer parameters during osmotic treatment, as well as it did the type of osmotic solution used. 32 Thermal analysis showed the variations in sugar melting temperature and enthalpy as a result of 33 osmotic treatment and storage. During the storage, a lower weight loss, and higher lightness was 34 observed in US pre-treated samples. The sample that preserved the best chemico-physical and 35 36 microbiological characteristics during storage was the one treated with 61.5% sucrose solution, due to its lowest water activity. 37

38

39 Keywords: cranberry, osmotic dehydration, shelf life, microbiology, water activity, colour

2

#### 40 1. Introduction

Cranberries are considered as a rich source of many compounds which have a positive impact on 41 human health. This fruit contains vitamins (A, C, E), minerals (potassium, sodium, selenium), fiber, 42 lutein and beta-carotene. However, the most represented substances in cranberry fruit are polyphenols 43 (Blumberg et al., 2013; McKay, Chen, Zampariello, & Blumberg, 2015; Neto, 2007; Nowacka, 44 Fijalkowska, Dadan, et al., 2018; Teleszko, 2011). Unfortunately, fresh cranberries have a very sour 45 and tart flavor that makes them rarely eatable as raw fruits. Although sugar is generally not desirable 46 in the diet, according to American nutritional recommendations, its use is applied to improve the taste 47 of products with highly valuable nutritional values to make them acceptable for the consumer 48 (Blumberg et al., 2013; Kowalska & Olejnik, 2016; Nowacka et al., 2017a). Recently, different 49 substances such as sweeteners or sweetness enhancer were proposed as an alternative to the use of 50 sucrose. One of them is steviol glycoside, which is approximately 300 times sweeter than sucrose and 51 it is highly appreciated for its technological purpose, since it is temperature and pH stable during the 52 processing and storage (Periche, Castelló, Heredia, & Escriche, 2015). 53

To obtain appropriate sweetness of the final product, fresh cranberries are subjected to osmotic 54 dehydration (OD) process (Nowacka, Fijalkowska, Wiktor, et al., 2018). OD is a widely used process 55 carried out in hypertonic solution, usually sugars. During OD, a mass exchange occurs, in particular, 56 57 water flows out from the fresh tissue into the surrounding solution and the osmotic substance enters the dehydrated tissue. The result is a partial dewatering impregnation, which allows an increase of 58 the shelf-life of OD material (Goula, Kokolaki, & Daftsiou, 2017; Kaymak-Ertekin, F. & Sultanolu, 59 2000; Nowacka, Śledź, Wiktor, & Witrowa-Rajchert, 2014; Radojčin et al., 2015; Rząca, Witrowa-60 Rajchert, Tylewicz, & Rosa, 2009; Tylewicz et al., 2011). However, this process is long and often 61 requires the acceleration of mass transfer using traditional methods as agitation and rotation or new 62 techniques as for example ultrasound (Deng & Zhao, 2008; Fernandes, Oliveira, & Rodrigues, 2008; 63 Goula et al., 2017; Nowacka, Tylewicz, Laghi, Dalla Rosa, & Witrowa-Rajchert, 2014). Ultrasound 64 (US) is an air vibration in the frequency from 20 kHz to 100 kHz. In liquid medium it results in 65

compression and expansion of material, called "sponge effect", and formation of cavitation bubbles.
The application of ultrasound may have an influence on physical and chemical properties of plant
tissue due to creation of microscopic channels and formation of free radicals (Goula et al., 2017;
Knorr, Zenker, Heinz, & Lee, 2004; McClements, 1995; Nowacka, Fijalkowska, Dadan, et al., 2018;
Simal, Benedito, Sánchez, & Rosselló, 1998; Witrowa-Rajchert, Wiktor, Sledz, & Nowacka, 2014).
Moreover, ultrasound coupled with thermal treatment leads to better bacterial inactivation in
comparison to only thermal treatment (Zenker et al., 2003).

It is recognized that quality characteristics and physical properties of food may undergo changes 73 during US treatment (Fernandes, Gallão, & Rodrigues, 2009; Fernandes, Oliveira, et al., 2008; Goula 74 et al., 2017; Kentish & Ashokkumar, 2011; Nowacka, Fijalkowska, Dadan, et al., 2018; Rajewska & 75 Mierzwa, 2017) and OD process (Fernandes, Gallão, & Rodrigues, 2009; Nowacka, Tylewicz, 76 Romani, Dalla Rosa, & Witrowa-Rajchert, 2017b), but also during further storage (Wang, 2006). 77 However, while changes due to process have received more attention, evolution of quality during 78 storage of these type of product are seldom assessed. Thus, the aim of this study was to investigate 79 the effect of ultrasound assisted osmotic dehydration on cranberries mass exchange parameters and 80 on their quality parameters (dry matter, water activity, colour, thermal behaviour and microbial 81 growth) during storage. 82

83

## 84 2. Material and methods

## 85 2.1. Sample preparation

Fresh swamp cranberry fruits (*Vaccinium oxycoccus*) were used in this research. The fruits were bought on the Polish market and stored at  $4 \pm 1$ °C until processing. The cranberries were cut with a sharp knife in the geometric centre of the fruit to break hard skin and consequently to facilitate mass transfers during processes (Nowacka et al., 2017a). Fruits were subjected to ultrasound treatment and then to osmotic dehydration process.

91

#### 92 **2.2.** Sonication procedure (US)

50g of samples were placed in a beaker into OD solutions in ratio of 1:4 (fruit:solution) (Fernandes, 93 Gallão, & Rodrigues, 2008; Tylewicz et al., 2011) in order to avoid changes in the solution 94 concentration and then positioned in an ultrasonic bath MKD-3 (MKD Ultrasonics, Stary Konik, 95 Poland, internal dimensions: 240x140x110 mm). Sonication was conducted for 30 minutes 96 (Nowacka, Fijalkowska, Wiktor, et al., 2018) using frequency 21 kHz and the total power generated 97 by sonotrodes 180 W, that corresponded to an intensity of 3.6 W/g. During treatments the fruits were 98 covered by a net in order to prevent them from flowing to the surface. During the sonication in OD 99 solutions, significant temperature changes  $(\pm 1^{\circ}C)$  were not observed. The treatment was conducted 100 101 in two repetitions for each osmotic solution. After the sonication, beakers were transferred to a rotary shaker with controlled temperature to continue the osmotic dehydration process. 102

103

#### 104 **2.3.** Osmotic dehydration (OD)

105 Osmotic dehydration was carried out in three different solutions at the temperature of 40°C:

106 1. 61.5% sucrose solution (SA)

#### 107 2. 30% sucrose solution with 0.1% of steviol glycosides (STV)

108 3. 40% trehalose solution (T).

109 SA solution is usually used for osmotic dehydration as standard solution (Ciurzyńska, Kowalska, Czajkowska, & Lenart, 2016). The solution of 30% of sucrose with addition of natural sweetener as 110 steviol glycosides was used in order to reduce the sucrose content and keep similar sweetness. Steviol 111 glycosides are up to 300 times sweeter than sucrose, thus the addition of 0.1% of steviol glycosides 112 to a 30% sucrose solution provide a comparable sweetness to a 61.5% sucrose solution (Nowacka et 113 al., 2017a). Trehalose solution was used as osmotic agent since it is able to improve the food structure 114 and contribute to microbiological stability (Dermesonlouoglou, Zachariou, Andreou, & Taoukis, 115 2016). Solutions were prepared by dissolving the solutes into distilled water. The sucrose (Pfeifer & 116 Langen Marketing Inc.) was used to prepare standard solutions. The steviol glycosides (Hortimex 117

Plus Inc.) with purity of 95,48% (63.43% of rebaudioside A, 22.85% of stevioside, 8.21% of
rebaudioside C, 0.73% of dulcoside A and 0.26% of steviolbioside) and trehalose (Exacta+Optech
Labcenter S.P.A) were used.

Due to high acidity of swamp cranberries, in order to obtain an acceptable taste of the product, the OD process was carried out till 72h of immersing in osmotic solution (Nowacka, Fijalkowska, Dadan,

123 et al., 2018). The samples subjected to OD process obtained a sweet-sour taste, which is characteristic

124 for processed cranberry fruits available on the market as jams, sauces or dried fruits.

After the treatment, the cranberries were rinsed with distilled water for 10 seconds and dried with absorbent paper for 5 seconds. The experiment was performed in duplicate for each solution. All

examined samples are summarized in Table 1.

128

### 129 **2.4.** Storage conditions

After osmotic dehydration, samples were packed in micro-perforated plastic bags made from polylactid acid (PLA) and stored for 8 weeks in a climatic chamber at the temperature of 10°C. The chosen quality properties were analysed immediately after OD treatment (T0) and after the first (T1), second (T2), fourth (T4) and eighth (T8) week of storage. For each storage time two bags for each sample were analysed.

135

# 136 **2.5.** Analytical determinations

## 137 **2.5.1. Mass transfer parameters**

The mass transfer after 72 h of OD process was evaluated in terms of weight reduction (WR, kg·kg<sup>-1</sup>), water loss (WL, kg·kg<sup>-1</sup>) and solid gain (SG, kg·kg<sup>-1</sup>) according to the following equations (Tylewicz et al., 2017):

141 
$$WR = \frac{m_t - m_0}{m_0}$$
(1)

142 
$$WL = \frac{m_t x_{wt} - m_0 x_{w0}}{m_0}$$
(2)

)

143 
$$SG = \frac{m_t x_{STt} - m_0 x_{ST0}}{m_0}$$
(3)

144

- 145 where:
- 146  $m_0$  initial weight before osmotic treatment (kg)
- 147  $m_t$  weight after a time t (kg)
- 148  $x_{w0}$  initial water mass fraction (kg·kg<sup>-1</sup>)
- 149  $x_{wt}$  water mass fraction after a time t (kg·kg<sup>-1</sup>)
- 150  $x_{ST0}$  initial total solids (dry matter) mass fraction (kg·kg<sup>-1</sup>)
- 151  $x_{STt}$  total solids (dry matter) mass fraction after a time t (kg·kg<sup>-1</sup>)
- 152 Moreover, weight reduction was also calculated for all the samples during the storage of 8 weeks,
- taking into account the weight of the samples at T0 and at each time of storage.
- 154

## 155 **2.5.2. Moisture content**

- Moisture content was determined gravimetrically by drying the samples at 70°C until a constant weight was achieved (AOAC, 2002).
- 158

### 159 **2.5.3. Water activity**

160 Water activity (a<sub>w</sub>) was determined using the device AquaLab Series 3TE (Decagon Devices Inc.,

161 Pullman, USA). The assay was performed in triplicate at room temperature  $(23\pm1^{\circ}C)$ .

162

#### 163 **2.5.4.** Colour

Colour was analysed using a spectro-photocolorimeter (HUNTERLAB Color-FlexTM, A60-1010-615, Reston, Virginia) equipped with a 12 mm diameter sample holder. The following parameters were used: illuminant D65 (6500 K), observer 10°. Before each series of measurements, the instrument was calibrated with a black and white tile (L\* = 93.47; a \* = -0.83; b \* = 1.33). The colour data were expressed, according to the CIE L\*a\*b\* scale, in L\* (lightness), a\* (index of red) and b\* (index of yellow). The final values were calculated as an average of nine measures. Moreover, the

hue angle (h°) and the total colour differences ( $\Delta E$ ) were calculated with the equation 4 and 5 respectively (Radojčin et al., 2015; Wiktor, Sledz, Nowacka, Rybak, & Witrowa-Rajchert, 2016):

172 
$$h^{o} = tan^{-1}\frac{b^{*}}{a^{*}}$$
 (4)

173 
$$\Delta E = \sqrt{\left(\Delta L^*\right)^2 + \left(\Delta a^*\right)^2 + \left(\Delta b^*\right)^2}$$

174 where:

175  $\Delta L^*$ ,  $\Delta a^*$ ,  $\Delta b^*$  is the differences of mean L\*,  $a^*$  and  $b^*$  parameters, respectively, between fresh 176 cranberries and osmodehydrated untreated and US treated samples.

(5)

177

# 178 **2.5.5. Differential Scanning Calorimetry measurements (DSC)**

The calorimetric analysis was performed to determine the temperature of sugars' decomposition using 179 a DSC Q20 model differential calorimeter (TA Instrument, Germany) according to Panarese, 180 Tylewicz, Santagapita, Rocculi, & Dalla Rosa (2012). The DSC was equipped with a cooling unit 181 182 (TA-Refrigetated Cooling System90). The calibration of temperature and melting enthalpies was carried out with distilled water (T<sub>m</sub> 0.0 °C) and indium (T<sub>m</sub> 156.60 °C). For the calibration and for 183 sample measurements the same heating rate was used, under a 50 mL/min dry nitrogen flow. About 184 20-30 mg of each sample were weighed in 50 µl aluminum capsules, closed and punctured just before 185 the measurement. An empty capsule was used as a reference. The curves were obtained by heating 186 the samples from 20 to 300 °C at a rate of 10 °C/min. 187

188

#### 189 **2.5.6.** Microbiology assessment

The microbiological analyses were performed immediately after the osmotic treatment conducted for 72 hours (T0) and during the storage period (after 1, 2, 4, 8 weeks). In particular, the cell loads of mesophilic aerobic bacteria, yeasts and moulds were monitored in all samples over the storage, according to the procedure reported by Mannozzi et al. (2018). The results are the average of three independent samples for each condition. 195

#### 196 2.6. Statistical analysis

197 Analysis of variance (ANOVA) was performed using Statistical 7.0 Statsoft software (Tulsa, UK) 198 using the Duncan test with a level of p < 0.05.

199

#### 200 **3. Results and discussion**

#### **3.1. Mass transport balance during the OD treatment**

Table 2 shows the results of mass transfer parameters (weight reduction, water loss, solid gain) 202 involved during the OD treatment for 72 h at 40 °C. From the table it is possible to observe that the 203 204 highest weight reduction was observed for samples treated with sucrose (SA), followed by samples treated with sucrose in combination with steviol glycoside (STV) and samples dehydrated with 205 trehalose (T). These differences were similar to the ones observed for the water loss from the samples. 206 In fact, the highest water loss was observed for samples treated with high concentration of sucrose. 207 This is due to the difference in osmotic pressure between food matrix and osmotic solution. SA 208 solution presented the higher concentration of sugar (61.5%) and was characterized by the lowest a<sub>w</sub> 209 of about 0.867, while the solutions of STV and T presented the values of 0.976 and 0.965, 210 respectively. 211

In a study investigating the dehydration of apples, when sucrose and trehalose were used as osmotic solution at the same  $a_w$  (0.96) lower water mass fraction (0.819 g  $\cdot$  g<sup>-1</sup>) was observed in samples dehydrated with sucrose solution against 0.838 g  $\cdot$  g<sup>-1</sup> observed for trehalose solution, indicating the lower dewatering for the latest samples (Atarés, Chiralt, Corradini, & González-Martínez, 2009). Also Tylewicz et al. (2017) observed that samples treated with trehalose solution at the same concentration as sucrose (40% w/w) were characterized by a lower final dehydration level (at 120 min of OD) even if a higher initial rate of dehydration was observed.

As explained by Galmarini et al. (2011) at the same solute concentration, the  $a_w$  (which explains the

engine of transport) is lower in sucrose solution as well as its viscosity (which explains the molcular

diffusion). Therefore, the water transport is faster in samples dehydrated in sucrose than these dehydrated with trehalose.

223 Concerning the solid gain, the highest value was observed in samples treated with sucrose, followed 224 by the samples treated with trehalose. Trehalose was found to have a higher effect on solid gain when 225 used at the same concentration as sucrose (Tylewicz et al., 2017). In the present study, while a slightly 226 but significantly higher water loss was observed for STV samples compared to T samples, the latter 227 showed almost twice the solid gain.

Ultrasound pre-treatment led to a significant increase of weight reduction and water loss in all the samples. It is well known that ultrasound create microscopic channels which may ease moisture removal and increase the diffusivity of the water (Fernandes, Gallão, et al., 2008; Fernandes & Rodrigues, 2007; Nowacka et al., 2014). On the other side, for SA\_US and T\_US samples the US pre-treatment did not promote any differences for solid gain, while it caused a significant decrease in samples STV\_US. This is probably because the solid gain comprises both soluble and insoluble solids (fruit matrix), and therefore any lysis effect in the tissue is accounted in this group.

235

#### **3.2. Weight reduction and dry matter content during storage**

Weight reduction and dry matter content of osmodehydrated cranberries throughout storage are 237 shown respectively in Figure 1 and 2. Weight loss was generally in the range of 2-5%. The US pre-238 treatment led to a significantly lower weight loss in samples treated with SA and STV in comparison 239 to the samples only osmodehydrated. The samples treated with sucrose solution both with or without 240 241 US application presented the lowest weight loss, followed by samples treated by trehalose and then the samples treated with sucrose and steviol glycoside, which presented the highest weight loss. In 242 general, the loss of weight during storage could be due to the water loss that resulted from surface 243 water evaporation, and also by gravity mechanism that moves the liquid phase to the bottom of 244 samples, leaving the tissue. Moreover, respiration and transpiration as well as the wounding stress 245 during the processing of the fruits could cause the higher weight gain during the storage (Ferrari, 246

Sarantópoulos, Carmello-Guerreiro, & Hubinger, 2013). In fact, in the case of our study the weight loss was the highest in samples dehydrated with STV, which probably could still maintain the biological capacity to the cell respiration or even to ferment. Due to the higher dehydration during the process, samples treated with sucrose presented also the lower water content (Figure 2) compared to the other samples, that could be the reason for the lower loss of water during the storage.

As shown in Figure 2, the highest value of dry matter was observed for cranberries samples treated 252 at 61.5% sucrose solution (around 0.5%). As already observed for mass transfer parameters, these 253 samples were subjected to a higher dewatering and thus higher increase of the dry mater. During 254 storage, a further (even if only slight) increase of dry mater was observed in all samples. Higher dry 255 matter observed in SA US and STV US treated samples, in comparison to their controls, 256 immediately after the treatment and at different period of storage was probably due to the partial 257 destruction of the fruit peel allowing a higher penetration of sucrose into the tissue (Nowacka et al., 258 2017a). 259

260

#### 261 **3.3. Water activity**

Figure 3 shows the water activity evolution for osmodehydrated cranberry samples with or without 262 US pre-treatment during storage. The samples treated with SA presented the lowest a<sub>w</sub> in comparison 263 264 to the samples dehydrated with other solutions, because of the higher osmotic potential differences. The application of US did not change significantly the water activity of the samples at T0. Also, 265 Stojanovic and Silva (2006) and Nowacka et al. (2017b) did not observed differences in water activity 266 of ultrasound osmodehydrated blueberry and kiwifruit samples, respectively. An increase of water 267 activity was observed at T1, while during further storage a slight but progressive decrease of water 268 activity was noticed. The results are in accordance with the dry matter content, in fact the increase of 269 solutes content during storage could lead to the decrease of water activity. 270

271

#### 272 **3.4. Colour changes during storage**

11

Table 3 shows the changes of colour parameters of lightness (L\*), red index (a\*), yellow index (b\*), 273 hue angle (h°) and total colour differences ( $\Delta E$ ). During storage, until the fourth week a decrease of 274 the L\* parameter was observed for all samples. Cranberries dehydrated with ultrasound in trehalose 275 solution (T US) showed the lowest lightness value. Obtained data are in agreement with those 276 reported in the literature, according to which the osmotic treatment leads to a decrease of the initial 277 L\* values. This phenomenon can be related to the modification of the surface laver of the product 278 due to immersion in sugary solutions. This effect was noticed by Prinzivalli et al. (2006) in strawberry 279 slices. As reported in the literature in the case of dehydrated apples and carrots, the pre-treatment 280 with ultrasound leads to an increase in lightness (L\*) compared to the untreated fruit (Fijalkowska, 281 Nowacka, Wiktor, Sledz, & Witrowa-Rajchert, 2016). In our case, significantly higher values of L\* 282 for untreated and US treated samples were observed only at the 8<sup>th</sup> week of storage. Probably these 283 colour changes were associated with loss of the anthocyanins during storage, which are responsible 284 285 for red colour of cranberry fruits (Nowacka, Fijalkowska, Dadan, et al., 2018; Oszmiański, Wojdyło, Lachowicz, Gorzelany, & Matłok, 2016). This was also confirmed by the changes of a\* parameter, 286 which represent the red colour. In general, a decrease of a\* and b\* parameters was noted during 287 storage, while the hue angle values, in the range of 16 to 28, indicating a red hue of the samples, did 288 not show a real trend. In fact, they first decreased after one week and then started to increase till the 289 290 fourth week, and then again decreased at eight week of storage.

In order to evaluate the colour alteration, the total colour differences was calculated. Generally, the 291 value of  $\Delta E$  higher than 2 is considered as a visible difference between the samples (Fijalkowska, 292 Nowacka, & Witrowa-Rajchert, 2017). All samples were characterized by high values of  $\Delta E$  in the 293 range from 6 to 10 after the osmotic dehydration process and the total colour differences grow during 294 storage till 4<sup>th</sup> week. In the 8<sup>th</sup> week of storage the decrease of  $\Delta E$  was observed. The changes of 295 colour are usually connected with the chemical compounds, which are responsible for the colour e.g. 296 anthocyanins (Nowacka, Fijalkowska, Dadan, et al., 2018). In particular, red colour is one of the main 297 indicators by which the consumer can assesses the quality of fresh cranberries. It makes the product 298

more or less attractive and it is the first characteristic that the consumer perceives (Kutyła-Olesiuk, Nowacka, Wesoły, & Ciosek, 2013; Nowacka, Fijalkowska, Wiktor, et al., 2018). The reduction of the red component of colour might be connected with the solubilization of pigments in the solution during osmotic treatment and the degradation of anthocyanins. Anthocyanins are particularly sensitive to osmotic dehydration, which may be the reason for the general reduction in colour (de Bruijn & Bórquez, 2014).

305

### **306 3.5. Thermal analysis DSC results**

307 DSC measurements were performed to evaluate the melting of sugars introduced by OD in the 308 samples and to evaluate any differences during storage. The melting temperature (Tm, °C) and 309 enthalpy ( $\Delta$ H, J·g<sup>-1</sup>) registered in the sample are reported in Figure 4 and Figure 5, respectively.

Melting of sucrose is known to occur in the temperature range of 206-232°C (Abd-Elrahman & Ahmed, 2009; Panarese, Tylewicz, Santagapita, Rocculi, & Dalla Rosa, 2012). In the present study, after the OD treatment a melting temperature of 211 °C was found, which is consistent with literature data previously reported.

The presence of steviol glycosides at 0.1% did not influence the melting temperature of the solution. In the case of trehalose, a melting peak with a temperature of 246 °C was observed. While about the crystallization of trehalose and its influence on glass transition there is a wide literature, its melting behaviour is not often studied. Raemy and Schweizer (1983) studied the melting properties of different sugars and indicated a melting temperature for trehalose of 235 °C, higher compared to sucrose one (215°C), which is similar to what we found in the present study.

320 No differences were observed during storage in the peaks of melting temperatures for all the samples.

321 The enthalpy values are found to be proportional to the sugar contents. The initial values reported in

322 Figure 5, of about 40, 27 and 19  $J \cdot g^{-1}$  respectively for SA, T and STV samples, reflect the

impregnation levels reported in Table 2 for samples after 72 h of OD process. While for STV sample,

no differences were observed when US was applied, for SA and T samples, an increase in the melting

enthalpy was observed. This difference may be attributed to the differences in mass transfer as shown
by Table 2. Indeed, while water loss increased in STV sample after US application (increase of 8%,
compared to 2 and 6% of respectively SA and T samples), solid gain decreased for of about 1.7%.
Moreover, it could be possible that T and T\_US samples suffer partial hydrolyzation of trehalose.

#### 330 **3.6. Microbial analysis**

In Table 4 the results of total mesophilic aerobic bacteria and yeasts in cranberries samples during 331 the shelf-life are reported. Osmotic dehydration improves the microbiological shelf-life since it leads 332 to a reduction in water activity (Castelló, Igual, Fito, & Chiralt, 2009). In fact, even in our case study, 333 the samples that reached lower water activity (SA; SA US30) showed a better microbiological shelf-334 life over time. This is probably because of the high osmotic pressure of the SA solution which can 335 enable longer shelf-life. Moreover, sucrose absorption has promoted an increase in the viscosity of 336 the liquid phase, influencing the kinetics of the microbial growth, as reported in the case of kiwifruit 337 by (Gianotti, Sacchetti, Guerzoni, & Dalla Rosa, 2001). According to the recommended 338 microbiological criteria for fruits and vegetables, the maximum limits for the total mesophilic aerobic 339 bacteria and yeasts/moulds are set to be 1 x 10<sup>4</sup> CFU/g and 1 x 10<sup>2</sup> CFU/g, respectively (Pascual and 340 Calderón, 2000). As reported in Table 4, there were no significant differences between the samples 341 pre-treated or not with ultrasound. However, the cell load of the total mesophilic aerobic bacteria, 342 exceeded the maximum levels recommended by Pascual and Calderón (2000), after the first week of 343 storage for the samples osmodehydrated in solutions with 30% sucrose + 0.1% stevia and 40%344 trehalose that were subjected to ultrasound pre-treatment. During the second week of storage this 345 microbiological limit was also exceeded for samples osmodehydrated with sucrose + stevia and 346 trehalose and not subjected to ultrasound pre-treatment, whereas for samples osmodehydrated with 347 61.5% of sucrose solution, with and without US pre-treatment, this limit was only exceeded after 348 eighth weeks of storage. As regard to yeasts, the recommended cell load was superseded by all the 349 samples at the beginning of the first week of storage, whereas for moulds the cell load exceeds only 350

for samples osmodehydrated with trehalose. During the fourth week of storage, the recommended cell load for moulds 1 x  $10^2$  CFU/g was superseded by all samples with the exception of those osmodehydrated with 61.5% sucrose that exceeded this limit only during the eighth week (data not shown) of storage. In fruits and vegetables, intercellular spaces play an important role in the penetration of microorganisms. In general, bacteria, yeast or mould cells are much smaller than plant cells, thus they can easily penetrate into the vegetable tissues (Alzamora et al., 2005).

357

## 358 Conclusions

The mass transfer parameters during OD treatment were significantly influenced by ultrasound 359 application and the type of osmotic solution used. The highest water loss was observed in samples 360 treated with sucrose and ultrasound (SA US). During storage, ultrasound pre-treatment led to lower 361 weight reduction in cranberry samples treated with any type of the solution in comparison to those 362 without US pre-treatment. Moreover, during storage, the US pre-treatment promoted changes in the 363 qualitative characteristics, in particular of colour leading to a higher lightness (L\*) in comparison to 364 the untreated fruit. The thermal analysis allowed the identification of the variations in melting 365 temperatures and enthalpies as a result of osmotic treatment and storage. 366

The sample that preserved the best chemico-physical and microbiological characteristics during storage was the one treated with 61.5% sucrose solution, due to the lowest water activity. However, further studies are necessary in order to better understand the chemico-physical and physiological mechanisms underlying the highlighted changes.

371

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375

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#### 543 **Figure Caption**

Figure 1. Weight reduction of osmodehydrated cranberries with or without US pre-treatment during storage at 10 °C. Different letters indicate statistical differences (p < 0.05) (lowercase letter within the same row indicate differences among different treatments for the same storage time, while capital letters within the same column indicate differences among different storage times for the same treatment).

Figure 2. Dry matter content ( $g \cdot g^{-1}$ ) of osmodehydrated cranberries with or without US pre-treatment during storage at 10 °C. Different letters indicate statistical differences (p < 0.05) (lowercase letter within the same row indicate differences among different treatments for the same storage time, while capital letters within the same column indicate differences among different storage times for the same treatment).

Figure 3. Water activity of osmodehydrated cranberries with or without US pre-treatment during storage at 10 °C. Different letters indicate statistical differences (p < 0.05) (lowercase letter within the same row indicate differences among different treatments for the same storage time, while capital letters within the same column indicate differences among different storage times for the same treatment).

Figure 4. The melting temperature (Tm, °C) of osmodehydrated cranberries with or without US pretreatment during storage at 10 °C. Different letters indicate statistical differences (p < 0.05) (lowercase letter within the same row indicate differences among different treatments for the same storage time, while capital letters within the same column indicate differences among different storage times for the same treatment).

Figure 5. Melting enthalpy ( $\Delta$ H, J·g<sup>-1</sup>) of osmodehydrated cranberries with or without US pretreatment during storage at 10 °C. Different letters indicate statistical differences (p < 0.05) (lowercase letter within the same row indicate differences among different treatments for the same storage time, while capital letters within the same column indicate differences among different storage times for the same treatment).



Time (weeks)









Abbreviations	Sample description				
SA	cranberries subjected to osmotic dehydration with 61.5% sucrose solution				
STV	cranberries subjected to osmotic dehydration with 30% sucrose solution with				
	addition 0.1% of steviol glycosides				
Т	cranberries subjected to osmotic dehydration with 40% trehalose solution				
SA_US	cranberries subjected to 30 min ultrasound treatment and then osmotic				
	dehydration with 61.5% sucrose solution				
STV_US	cranberries subjected to 30 min ultrasound treatment and then osmotic				
	dehydration with 30% sucrose solution with addition 0.1% of steviol glycosides				
T_US	cranberries subjected to 30 min ultrasound treatment and then osmotic				
	dehydration with 40% trehalose solution				

Table 1. Abbreviations of examined cranberry samples

Table 2. Weight reduction  $(kg \cdot kg^{-1})$ , water loss  $(kg \cdot kg^{-1})$ , solid gain  $(kg \cdot kg^{-1})$  of osmodehydrated cranberries with or without US pre-treatment after 72 h of osmotic dehydration in sucrose (SA) sucrose + steviol glycoside (STV) and trehalose (T) solutions

Samples	Weight reduction	Water loss	Solid gain
SA	$0.402 \pm 0.002^{b}$	$0.586 \pm 0.002^{b}$	$0.184 \pm 0.002^{a}$
STV	$0.234\pm0.001^{\text{d}}$	$0.332 \pm 0.001^{\circ}$	$0.098 \pm 0.001^{\circ}$
Т	$0.172\pm0.001^{\rm f}$	$0.328\pm0.001^{\text{f}}$	$0.156 \pm 0.001^{b}$
SA_US	$0.424 \pm 0.002^{a}$	$0.605 \pm 0.002^{a}$	$0.182 \pm 0.002^{a}$
STV_US	$0.299 \pm 0.001^{\circ}$	$0.381 \pm 0.001^{\circ}$	$0.081 \pm 0.001^{d}$
T_US	$0.189\pm0.001^{\rm e}$	$0.345\pm0.001^{\text{d}}$	$0.156\pm0.001^{b}$

Different letters within the same column indicate statistical differences (p < 0.05).

L*					
	Т0	T1	T2	T4	Т8
SA	21 ± 3 <sup>aA</sup>	$17 \pm 1$ aAB	$17 \pm 1$ aAB	$15 \pm 1$ <sup>abB</sup>	$24 \pm 3 \text{ abA}$
STV	$17 \pm 1$ bB	16.6±0.4 <sup>aB</sup>	$15 \pm 1$ <sup>aB</sup>	$15 \pm 2 \ ^{abB}$	$21.8\pm0.1~^{\text{bA}}$
Т	$18 \pm 2 \ ^{abB}$	$18 \pm 2 \text{ aAB}$	$15 \pm 3 \ ^{aB}$	$13 \pm 3 \text{ abB}$	$22 \pm 1$ <sup>abA</sup>
SA_US	$20\pm3~^{abB}$	$18 \pm 1 \ ^{aB}$	$17\pm2~^{aB}$	$16\pm2^{aB}$	$25 \pm 1$ aA
STV_US	$18 \pm 2 \ ^{abB}$	$17 \pm 1$ <sup>aB</sup>	$19 \pm 3 \text{ aAB}$	$14 \pm 3 \text{ abB}$	$22.2 \pm 0.3 \text{ abA}$
T_US	$19\pm2~^{abB}$	$19\pm1~^{aBC}$	$15 \pm 2 \ ^{aC}$	$10.2 \pm 0.5 ^{bD}$	$23 \pm 1 \ ^{abA}$
a*					
	Т0	T1	T2	Τ4	Τ8
SA	$26\pm2^{aA}$	$24 \pm 1$ <sup>aA</sup>	$24 \pm 2 aA$	$28 \pm 3 \text{ aA}$	$18 \pm 1 \ ^{abB}$
STV	$25 \pm 2 \ ^{abA}$	$21 \pm 1 \ ^{aBC}$	$22 \pm 2 \ ^{aAB}$	$24\pm2~^{aAB}$	$17.6 \pm 0.1$ <sup>abC</sup>
Т	$25\pm2^{abAB}$	$21 \pm 3 \ ^{aBC}$	$23\pm3~\text{aABC}$	$27\pm2~^{aA}$	$19 \pm 2$ <sup>aC</sup>
SA_US	$24\pm1~^{abB}$	$22\pm2~^{aB}$	$25\pm2{}^{aAB}$	$29\pm2{}^{aA}$	$18 \pm 1 \ ^{abC}$
STV_US	$23 \pm 2^{bA}$	$21 \pm 2 aA$	$22 \pm 2 \ ^{aA}$	$25\pm3$ <sup>aA</sup>	$15 \pm 1 \text{ bcB}$
T_US	$25 \pm 2^{abA}$	$23\pm2^{aAB}$	$20\pm1~^{aB}$	$28 \pm 1 \ ^{aA}$	$14 \pm 1 ^{\text{cC}}$
b*					
	ТО	T1	T2	T4	Т8
SA	$12 \pm 1  ^{aB}$	$10 \pm 1 \text{ aAB}$	$10 \pm 2 ^{aAB}$	$15 \pm 2 ^{aA}$	8.3±1 <sup>abC</sup>
STV	$10 \pm 1$ <sup>abAB</sup>	$6 \pm 1 \ ^{bC}$	$8\pm4{}^{aABC}$	$11 \pm 4 ^{aA}$	$6.4 \pm 0.4$ bcdBC
Т	$11 \pm 2^{abAB}$	$6\pm 2^{bC}$	$8 \pm 1 \ ^{aBC}$	$13 \pm 1 \ ^{aA}$	$7 \pm 1$ abcBC
SA_US	$11\pm1~^{abB}$	$9\pm1~^{abBC}$	$11 \pm 1 \text{ aBC}$	$15 \pm 1 \ ^{aA}$	$9\pm1~^{aC}$
STV_US	$9\pm2$ bAB	$8 \pm 1 ^{abAB}$	$8\pm2^{aAB}$	$12 \pm 2 ^{aA}$	$5 \pm 1 ^{cdB}$

Table 3. Colour parameters L\*, a\*, b\*, hue angle h° and total colour differences ( $\Delta E$ ) of osmodehydrated cranberries with or without US pre-treatment during storage storage at 10 °C

T_US	$10\pm3~^{abA}$	$9\pm1~^{abAB}$	$8\pm1~^{aAB}$	$12 \pm 2 \ ^{aA}$	$4.6\pm0.2~^{dB}$
h°					
	T0	T1	T2	T4	Τ8
SA	$25 \pm 2 \ ^{aB}$	$23 \pm 1 \ ^{aB}$	$23 \pm 1 \ ^{aB}$	$28 \pm 2 ^{aA}$	$24 \pm 2 \ ^{abAB}$
STV	$21 \pm 2 ^{abAB}$	$16\pm2^{aB}$	$19\pm1~^{aAB}$	$23\pm 6^{aA}$	$20 \pm 1 ^{cdAB}$
Т	$23\pm3~^{abA}$	$17 \pm 5 \ ^{aB}$	$18\pm2^{aAB}$	$25 \pm 2 aA$	$21.6\pm0.1~^{bcAB}$
SA_US	$25 \pm 2^{bAB}$	$22 \pm 2 \ ^{aB}$	$23\pm3~^{aAB}$	$27.9 \pm 0.3$ aA	$25 \pm 1 \text{ aAB}$
STV_US	$21\pm3~^{abA}$	$21 \pm 2 ^{aA}$	$20 \pm 4 \ ^{aA}$	$24 \pm 2 \ ^{aA}$	$19 \pm 1 \text{ cdA}$
T_US	$21 \pm 3 \text{ bA}$	$21 \pm 2^{aA}$	$21 \pm 1$ <sup>aA</sup>	$24 \pm 3$ aA	$18 \pm 1 ^{dA}$
ΔΕ					
	Τ0	T1	T2	T4	Τ8
SA	$6 \pm 2^{bC}$	8 ± 1 <sup>bC</sup>	$8 \pm 2^{aC}$	$21.3\pm0.3~^{\mathrm{aA}}$	$14 \pm 1 ^{cB}$
STV	$8\pm2~^{abC}$	$13 \pm 2 aB$	$11 \pm 2 aB$	$22.0\pm0.6~^{aA}$	$14.9\pm0.2~^{bcB}$
Т	$8 \pm 3 \text{ abC}$	$13 \pm 2 aB$	$11 \pm 2 aBC$	$21.0\pm0.4~^{aA}$	$14 \pm 2 ^{cB}$
SA_US	$8 \pm 1 \ ^{abC}$	$10 \pm 2^{abC}$	$7 \pm 3 \ ^{aC}$	$21.1\pm0.1~^{\mathrm{aA}}$	$14 \pm 1 \text{ bcB}$
STV_US	$10 \pm 3 \text{ aB}$	$11 \pm 2 \text{ abB}$	$11 \pm 3 \text{ aB}$	21 ± 1 <sup>aA</sup>	$17 \pm 2 \text{ abA}$
T_US	$8 \pm 2 \ ^{abC}$	$9 \pm 1$ abC	$13 \pm 2$ <sup>aB</sup>	$21.0\pm0.2~^{aA}$	$19 \pm 1$ <sup>aA</sup>

Different letters indicate statistical differences (p < 0.05) for each colour parameter (lowercase letter within the same column indicate differences among different treatments for the same storage time, while capital letters within the same row indicate differences among different storage times for the same treatment).

Mesophilic aerobic bacteria log CFU/g T0 T1 T2 T4 T8  $3.3\pm0.3^{b}$ SA  $2.2\pm0.2^{a}$  $2.9 \pm 0.2^{d}$  $2.9\pm0.3^{\text{c}}$  $4.9\pm0.2^{\circ}$ STV  $3.8\pm0.2^{b}$  $5.4 \pm 0.3^{\circ}$  $2.4\pm0.2^{a}$  $7.9\pm0.2^{a}$  $8.0\pm0.3^{a}$ Т  $2.0\pm0.3^{a}$  $4.8\pm0.2^{a}$  $6.0 \pm 0.2^{b}$  $7.2 \pm 0.2^{b}$  $8.2\pm0.2^{a}$  $2.5\pm0.3^{\text{a}}$  $1.5\pm0.3^{\circ}$  $2.8\pm0.2^{d}$  $2.9\pm0.2^{\text{c}}$  $5.5\pm0.2^{b}$ SA\_US STV US  $2.0\pm0.3^{a}$  $5.1 \pm 0.2^{a}$  $6.7 \pm 0.1^{a}$  $7.9\pm0.3^{a}$  $8.6 \pm 0.4^{a}$  $7.4\pm0.3^{ab}$  $5.2\pm0.3^{a}$  $T_US$  $2.5\pm0.2^{a}$  $7.0\pm0.3^{a}$  $8.4\pm0.3^{a}$ Yeast log CFU/g T0 T1 T2 T4 T8  $2.6 \pm 0.2^{a}$  $2.7\pm0.12^{\text{d}}$  $4.9\pm0.2^{b}$ SA  $1.5\pm0.2^{\circ}$  $1.9\pm0.5^{c}$ STV  $2.7\pm0.2^{a}$  $3.1 \pm 0.2^{b}$  $5.3 \pm 0.2^{\circ}$  $7.9\pm0.3^{a}$  $8.3\pm0.3^{\text{a}}$ 

 $6.1\pm0.2^{b}$ 

 $2.1 \pm 0.2^{e}$ 

 $6.6\pm0.2^{a}$ 

 $6.8\pm0.3^{a}$ 

 $7.1\pm0.2^{b}$ 

 $2.8\pm0.3^{\text{c}}$ 

 $7.8\pm0.3^{a}$ 

 $7.3 \pm 0.3^{ab}$ 

 $8.1\pm0.4^{a}$ 

 $5.2\pm0.2^{b}$ 

 $8.2\pm0.3^{a}$ 

 $8.3\pm0.3^{a}$ 

Table 4. Mesophilic aerobic bacteria and yeast of osmodehydrated cranberries with or without US pre-treatment during storage storage at 10 °C

Different letters within the same column indicate statistical differences (p < 0.05).

 $4.6 \pm 0.3^{a}$ 

 $1.0\pm0.4^{\circ}$ 

 $4.6 \pm 0.3^{a}$ 

 $5.2 \pm 0.3^{a}$ 

Т

SA-US

STV-US

T-US

 $1.9\pm0.2^{\text{c}}$ 

 $2.1\pm0.2^{bc}$ 

 $1.9 \pm 0.2^{\circ}$ 

 $2.5\pm0.2^{ab}$