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

3

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17

18

19 **Abstract**

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

38

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

40 1. Introduction

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

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

66 compression and expansion of material, called “sponge effect”, and formation of cavitation bubbles.
67 The application of ultrasound may have an influence on physical and chemical properties of plant
68 tissue due to creation of microscopic channels and formation of free radicals (Goula et al., 2017;
69 Knorr, Zenker, Heinz, & Lee, 2004; McClements, 1995; Nowacka, Fijalkowska, Dadan, et al., 2018;
70 Simal, Benedito, Sánchez, & Rosselló, 1998; Witrowa-Rajchert, Wiktor, Sledz, & Nowacka, 2014).
71 Moreover, ultrasound coupled with thermal treatment leads to better bacterial inactivation in
72 comparison to only thermal treatment (Zenker et al., 2003).
73 It is recognized that quality characteristics and physical properties of food may undergo changes
74 during US treatment (Fernandes, Gallão, & Rodrigues, 2009; Fernandes, Oliveira, et al., 2008; Goula
75 et al., 2017; Kentish & Ashokkumar, 2011; Nowacka, Fijalkowska, Dadan, et al., 2018; Rajewska &
76 Mierzwa, 2017) and OD process (Fernandes, Gallão, & Rodrigues, 2009; Nowacka, Tylewicz,
77 Romani, Dalla Rosa, & Witrowa-Rajchert, 2017b), but also during further storage (Wang, 2006).
78 However, while changes due to process have received more attention, evolution of quality during
79 storage of these type of product are seldom assessed. Thus, the aim of this study was to investigate
80 the effect of ultrasound assisted osmotic dehydration on cranberries mass exchange parameters and
81 on their quality parameters (dry matter, water activity, colour, thermal behaviour and microbial
82 growth) during storage.

83

84 **2. Material and methods**

85 **2.1. Sample preparation**

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

91

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

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

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

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

121 Due to high acidity of swamp cranberries, in order to obtain an acceptable taste of the product, the
 122 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.

125 After the treatment, the cranberries were rinsed with distilled water for 10 seconds and dried with
 126 absorbent paper for 5 seconds. The experiment was performed in duplicate for each solution. All
 127 examined samples are summarized in Table 1.

128

129 2.4. Storage conditions

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

135

136 2.5. Analytical determinations

137 2.5.1. Mass transfer parameters

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

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

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

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

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 ($\text{kg} \cdot \text{kg}^{-1}$)149 x_{wt} - water mass fraction after a time t ($\text{kg} \cdot \text{kg}^{-1}$)150 x_{ST0} - initial total solids (dry matter) mass fraction ($\text{kg} \cdot \text{kg}^{-1}$)151 x_{STt} - total solids (dry matter) mass fraction after a time t ($\text{kg} \cdot \text{kg}^{-1}$)

152 Moreover, weight reduction was also calculated for all the samples during the storage of 8 weeks,
153 taking into account the weight of the samples at T0 and at each time of storage.

154

155 **2.5.2. Moisture content**

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

158

159 **2.5.3. Water activity**

160 Water activity (a_w) was determined using the device AquaLab Series 3TE (Decagon Devices Inc.,
161 Pullman, USA). The assay was performed in triplicate at room temperature ($23 \pm 1^\circ\text{C}$).

162

163 **2.5.4. Colour**

164 Colour was analysed using a spectro-photocolorimeter (HUNTERLAB Color-Flex™, A60-1010-
165 615, Reston, Virginia) equipped with a 12 mm diameter sample holder. The following parameters
166 were used: illuminant D65 (6500 K), observer 10°. Before each series of measurements, the
167 instrument was calibrated with a black and white tile ($L^* = 93.47$; $a^* = -0.83$; $b^* = 1.33$). The colour
168 data were expressed, according to the CIE $L^*a^*b^*$ scale, in L^* (lightness), a^* (index of red) and b^*
169 (index of yellow). The final values were calculated as an average of nine measures. Moreover, the

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

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

$$173 \quad \Delta E = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2} \quad (5)$$

174 where:

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

177

178 2.5.5. Differential Scanning Calorimetry measurements (DSC)

179 The calorimetric analysis was performed to determine the temperature of sugars' decomposition using
 180 a DSC Q20 model differential calorimeter (TA Instrument, Germany) according to Panarese,
 181 Tylewicz, Santagapita, Rocculi, & Dalla Rosa (2012). The DSC was equipped with a cooling unit
 182 (TA-Refrigerated Cooling System90). The calibration of temperature and melting enthalpies was
 183 carried out with distilled water (T_m 0.0 °C) and indium (T_m 156.60 °C). For the calibration and for
 184 sample measurements the same heating rate was used, under a 50 mL/min dry nitrogen flow. About
 185 20-30 mg of each sample were weighed in 50 μ l aluminum capsules, closed and punctured just before
 186 the measurement. An empty capsule was used as a reference. The curves were obtained by heating
 187 the samples from 20 to 300 °C at a rate of 10 °C/min.

188

189 2.5.6. Microbiology assessment

190 The microbiological analyses were performed immediately after the osmotic treatment conducted for
 191 72 hours (T_0) and during the storage period (after 1, 2, 4, 8 weeks). In particular, the cell loads of
 192 mesophilic aerobic bacteria, yeasts and moulds were monitored in all samples over the storage,
 193 according to the procedure reported by Mannozi et al. (2018). The results are the average of three
 194 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

201 3.1. Mass transport balance during the OD treatment

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

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

219 As explained by Galmarini et al. (2011) at the same solute concentration, the a_w (which explains the
220 engine of transport) is lower in sucrose solution as well as its viscosity (which explains the molecular

221 diffusion). Therefore, the water transport is faster in samples dehydrated in sucrose than these
222 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.

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

235

236 3.2. Weight reduction and dry matter content during storage

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

247 Sarantópoulos, Carmello-Guerreiro, & Hubinger, 2013). In fact, in the case of our study the weight
248 loss was the highest in samples dehydrated with STV, which probably could still maintain the
249 biological capacity to the cell respiration or even to ferment. Due to the higher dehydration during
250 the process, samples treated with sucrose presented also the lower water content (Figure 2) compared
251 to the other samples, that could be the reason for the lower loss of water during the storage.
252 As shown in Figure 2, the highest value of dry matter was observed for cranberries samples treated
253 at 61.5% sucrose solution (around 0.5%). As already observed for mass transfer parameters, these
254 samples were subjected to a higher dewatering and thus higher increase of the dry matter. During
255 storage, a further (even if only slight) increase of dry matter was observed in all samples. Higher dry
256 matter observed in SA_US and STV_US treated samples, in comparison to their controls,
257 immediately after the treatment and at different period of storage was probably due to the partial
258 destruction of the fruit peel allowing a higher penetration of sucrose into the tissue (Nowacka et al.,
259 2017a).

261 3.3. Water activity

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

271

272 3.4. Colour changes during storage

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

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

299 more or less attractive and it is the first characteristic that the consumer perceives (Kutyła-Olesiuk,
300 Nowacka, Wesoły, & Ciosek, 2013; Nowacka, Fijalkowska, Wiktor, et al., 2018). The reduction of
301 the red component of colour might be connected with the solubilization of pigments in the solution
302 during osmotic treatment and the degradation of anthocyanins. Anthocyanins are particularly
303 sensitive to osmotic dehydration, which may be the reason for the general reduction in colour (de
304 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 (T_m , °C) and
309 enthalpy (ΔH , J·g⁻¹) registered in the sample are reported in Figure 4 and Figure 5, respectively.

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

314 The presence of steviol glycosides at 0.1% did not influence the melting temperature of the solution.
315 In the case of trehalose, a melting peak with a temperature of 246 °C was observed. While about the
316 crystallization of trehalose and its influence on glass transition there is a wide literature, its melting
317 behaviour is not often studied. Raemy and Schweizer (1983) studied the melting properties of
318 different sugars and indicated a melting temperature for trehalose of 235 °C, higher compared to
319 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·g⁻¹ respectively for SA, T and STV samples, reflect the
323 impregnation levels reported in Table 2 for samples after 72 h of OD process. While for STV sample,
324 no differences were observed when US was applied, for SA and T samples, an increase in the melting

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

329

330 **3.6. Microbial analysis**

331 In Table 4 the results of total mesophilic aerobic bacteria and yeasts in cranberries samples during
332 the shelf-life are reported. Osmotic dehydration improves the microbiological shelf-life since it leads
333 to a reduction in water activity (Castelló, Igual, Fito, & Chiralt, 2009). In fact, even in our case study,
334 the samples that reached lower water activity (SA; SA_US30) showed a better microbiological shelf-
335 life over time. **This is probably because of the high osmotic pressure of the SA solution which can**
336 **enable longer shelf-life. Moreover,** sucrose absorption has promoted an increase in the viscosity of
337 the liquid phase, influencing the kinetics of the microbial growth, as reported in the case of kiwifruit
338 by (Gianotti, Sacchetti, Guerzoni, & Dalla Rosa, 2001). According to the recommended
339 microbiological criteria for fruits and vegetables, the maximum limits for the total mesophilic aerobic
340 bacteria and yeasts/moulds are set to be 1×10^4 CFU/g and 1×10^2 CFU/g, respectively (Pascual and
341 Calderón, 2000). As reported in Table 4, there were no significant differences between the samples
342 pre-treated or not with ultrasound. However, the cell load of the total mesophilic aerobic bacteria,
343 exceeded the maximum levels recommended by Pascual and Calderón (2000), after the first week of
344 storage for the samples osmodehydrated in solutions with 30% sucrose + 0.1% stevia and **40%**
345 **trehalose** that were subjected to ultrasound pre-treatment. During the second week of storage this
346 microbiological limit was also exceeded for samples osmodehydrated with sucrose + stevia and
347 trehalose and not subjected to ultrasound pre-treatment, whereas for samples osmodehydrated with
348 61.5% of sucrose solution, with and without **US** pre-treatment, this limit was only exceeded after
349 **eight** weeks of storage. As regard to yeasts, the recommended cell load was superseded by all the
350 samples at the beginning of the first week of storage, whereas for moulds the cell load exceeds only

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

357

358 **Conclusions**

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

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

371

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375

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542

543 **Figure Caption**

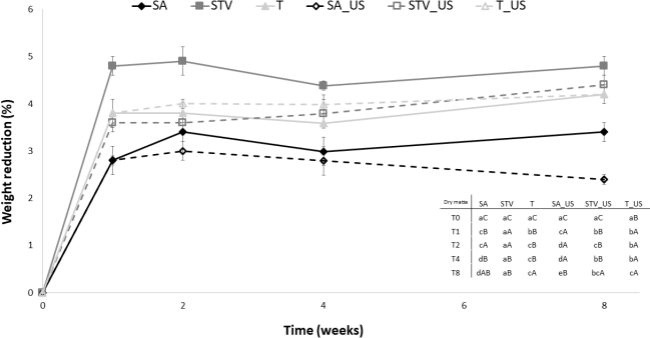
544 Figure 1. Weight reduction of osmodehydrated cranberries with or without US pre-treatment during
545 storage at 10 °C. Different letters indicate statistical differences ($p < 0.05$) (lowercase letter within
546 the same row indicate differences among different treatments for the same storage time,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

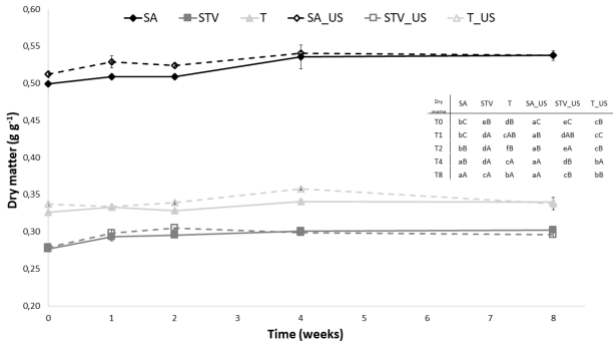
549 Figure 2. Dry matter content ($\text{g}\cdot\text{g}^{-1}$) of osmodehydrated cranberries with or without US pre-treatment
550 during storage at 10 °C. Different letters indicate statistical differences ($p < 0.05$) (lowercase letter
551 within the same row indicate differences among different treatments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

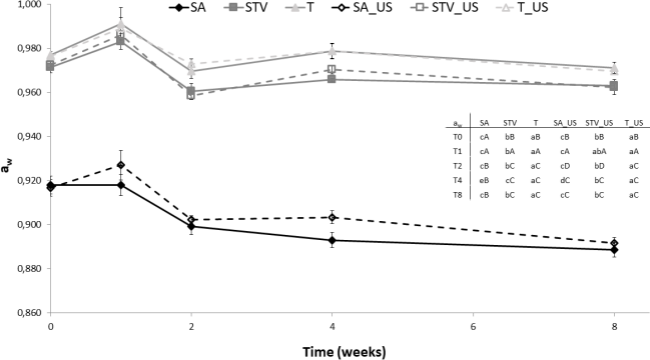
554 Figure 3. Water activity of osmodehydrated cranberries with or without US pre-treatment during
555 storage at 10 °C. Different letters indicate statistical differences ($p < 0.05$) (lowercase letter within
556 the same row indicate differences among different treatments for the same storage time,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

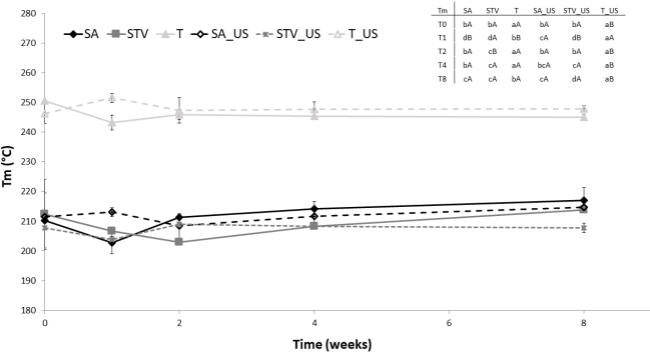
559 Figure 4. The melting temperature (T_m , °C) of osmodehydrated cranberries with or without US pre-
560 treatment during storage at 10 °C. Different letters indicate statistical differences ($p < 0.05$)
561 (lowercase letter within the same row indicate differences among different treatments for the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

564 Figure 5. Melting enthalpy (ΔH , $\text{J}\cdot\text{g}^{-1}$) of osmodehydrated cranberries with or without US pre-
565 treatment during storage at 10 °C. Different letters indicate statistical differences ($p < 0.05$)
566 (lowercase letter within the same row indicate differences among different treatments for the same
567 storage time, while capital letters within the same column indicate differences among different
568 storage times for the same treatment).









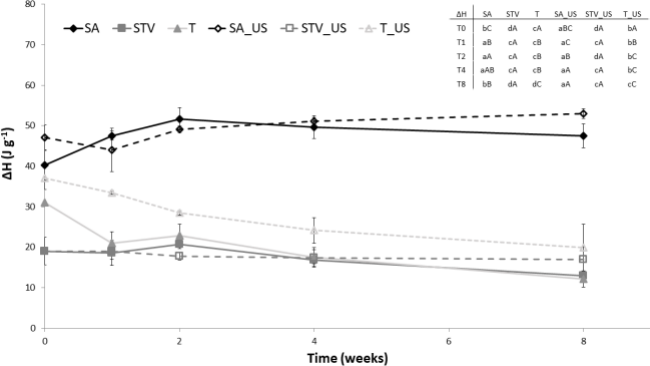


Table 1. Abbreviations of examined cranberry samples

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
T	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 2. Weight reduction ($\text{kg}\cdot\text{kg}^{-1}$), water loss ($\text{kg}\cdot\text{kg}^{-1}$), solid gain ($\text{kg}\cdot\text{kg}^{-1}$) 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 ± 0.002^b	0.586 ± 0.002^b	0.184 ± 0.002^a
STV	0.234 ± 0.001^d	0.332 ± 0.001^e	0.098 ± 0.001^c
T	0.172 ± 0.001^f	0.328 ± 0.001^f	0.156 ± 0.001^b
SA_US	0.424 ± 0.002^a	0.605 ± 0.002^a	0.182 ± 0.002^a
STV_US	0.299 ± 0.001^c	0.381 ± 0.001^c	0.081 ± 0.001^d
T_US	0.189 ± 0.001^e	0.345 ± 0.001^d	0.156 ± 0.001^b

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

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

L*					
	T0	T1	T2	T4	T8
SA	21 ± 3 ^{aA}	17 ± 1 ^{aAB}	17 ± 1 ^{aAB}	15 ± 1 ^{abB}	24 ± 3 ^{abA}
STV	17 ± 1 ^{bB}	16.6±0.4 ^{aB}	15 ± 1 ^{aB}	15 ± 2 ^{abB}	21.8 ± 0.1 ^{bA}
T	18 ± 2 ^{abB}	18 ± 2 ^{aAB}	15 ± 3 ^{aB}	13 ± 3 ^{abB}	22 ± 1 ^{abA}
SA_US	20 ± 3 ^{abB}	18 ± 1 ^{aB}	17 ± 2 ^{aB}	16 ± 2 ^{aB}	25 ± 1 ^{aA}
STV_US	18 ± 2 ^{abB}	17 ± 1 ^{aB}	19 ± 3 ^{aAB}	14 ± 3 ^{abB}	22.2 ± 0.3 ^{abA}
T_US	19 ± 2 ^{abB}	19 ± 1 ^{aBC}	15 ± 2 ^{aC}	10.2 ± 0.5 ^{bD}	23 ± 1 ^{abA}
a*					
	T0	T1	T2	T4	T8
SA	26 ± 2 ^{aA}	24 ± 1 ^{aA}	24 ± 2 ^{aA}	28 ± 3 ^{aA}	18 ± 1 ^{abB}
STV	25 ± 2 ^{abA}	21 ± 1 ^{aBC}	22 ± 2 ^{aAB}	24 ± 2 ^{aAB}	17.6 ± 0.1 ^{abC}
T	25 ± 2 ^{abAB}	21 ± 3 ^{aBC}	23 ± 3 ^{aABC}	27 ± 2 ^{aA}	19 ± 2 ^{aC}
SA_US	24 ± 1 ^{abB}	22 ± 2 ^{aB}	25 ± 2 ^{aAB}	29 ± 2 ^{aA}	18 ± 1 ^{abC}
STV_US	23 ± 2 ^{bA}	21 ± 2 ^{aA}	22 ± 2 ^{aA}	25 ± 3 ^{aA}	15 ± 1 ^{bcB}
T_US	25 ± 2 ^{abA}	23 ± 2 ^{aAB}	20 ± 1 ^{aB}	28 ± 1 ^{aA}	14 ± 1 ^{cC}
b*					
	T0	T1	T2	T4	T8
SA	12 ± 1 ^{aB}	10 ± 1 ^{aAB}	10 ± 2 ^{aAB}	15 ± 2 ^{aA}	8.3± 1 ^{abC}
STV	10 ± 1 ^{abAB}	6 ± 1 ^{bC}	8 ± 4 ^{aABC}	11 ± 4 ^{aA}	6.4± 0.4 ^{bcdBC}
T	11 ± 2 ^{abAB}	6 ± 2 ^{bC}	8 ± 1 ^{aBC}	13 ± 1 ^{aA}	7 ± 1 ^{abcBC}
SA_US	11 ± 1 ^{abB}	9 ± 1 ^{abBC}	11 ± 1 ^{aBC}	15 ± 1 ^{aA}	9 ± 1 ^{aC}
STV_US	9 ± 2 ^{bAB}	8 ± 1 ^{abAB}	8 ± 2 ^{aAB}	12 ± 2 ^{aA}	5 ± 1 ^{cdB}

T_US	10 ± 3 abA	9 ± 1 abAB	8 ± 1 aAB	12 ± 2 aA	4.6 ± 0.2 dB
h°					
	T0	T1	T2	T4	T8
SA	25 ± 2 aB	23 ± 1 aB	23 ± 1 aB	28 ± 2 aA	24 ± 2 abAB
STV	21 ± 2 abAB	16 ± 2 aB	19 ± 1 aAB	23 ± 6 aA	20 ± 1 cdAB
T	23 ± 3 abA	17 ± 5 aB	18 ± 2 aAB	25 ± 2 aA	21.6 ± 0.1 bcAB
SA_US	25 ± 2 bAB	22 ± 2 aB	23 ± 3 aAB	27.9 ± 0.3 aA	25 ± 1 aAB
STV_US	21 ± 3 abA	21 ± 2 aA	20 ± 4 aA	24 ± 2 aA	19 ± 1 cdA
T_US	21 ± 3 bA	21 ± 2 aA	21 ± 1 aA	24 ± 3 aA	18 ± 1 dA
ΔE					
	T0	T1	T2	T4	T8
SA	6 ± 2 bC	8 ± 1 bC	8 ± 2 aC	21.3 ± 0.3 aA	14 ± 1 cB
STV	8 ± 2 abC	13 ± 2 aB	11 ± 2 aB	22.0 ± 0.6 aA	14.9 ± 0.2 bcB
T	8 ± 3 abC	13 ± 2 aB	11 ± 2 aBC	21.0 ± 0.4 aA	14 ± 2 cB
SA_US	8 ± 1 abC	10 ± 2 abC	7 ± 3 aC	21.1 ± 0.1 aA	14 ± 1 bcB
STV_US	10 ± 3 aB	11 ± 2 abB	11 ± 3 aB	21 ± 1 aA	17 ± 2 abA
T_US	8 ± 2 abC	9 ± 1 abC	13 ± 2 aB	21.0 ± 0.2 aA	19 ± 1 aA

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).

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

Mesophilic aerobic bacteria log CFU/g					
	T0	T1	T2	T4	T8
SA	2.2 ± 0.2 ^a	3.3 ± 0.3 ^b	2.9 ± 0.2 ^d	2.9 ± 0.3 ^c	4.9 ± 0.2 ^c
STV	2.4 ± 0.2 ^a	3.8 ± 0.2 ^b	5.4 ± 0.3 ^c	7.9 ± 0.2 ^a	8.0 ± 0.3 ^a
T	2.0 ± 0.3 ^a	4.8 ± 0.2 ^a	6.0 ± 0.2 ^b	7.2 ± 0.2 ^b	8.2 ± 0.2 ^a
SA_US	2.5 ± 0.3 ^a	1.5 ± 0.3 ^c	2.8 ± 0.2 ^d	2.9 ± 0.2 ^c	5.5 ± 0.2 ^b
STV_US	2.0 ± 0.3 ^a	5.1 ± 0.2 ^a	6.7 ± 0.1 ^a	7.9 ± 0.3 ^a	8.6 ± 0.4 ^a
T_US	2.5 ± 0.2 ^a	5.2 ± 0.3 ^a	7.0 ± 0.3 ^a	7.4 ± 0.3 ^{ab}	8.4 ± 0.3 ^a
Yeast log CFU/g					
	T0	T1	T2	T4	T8
SA	2.6 ± 0.2 ^a	1.5 ± 0.2 ^c	2.7 ± 0.12 ^d	1.9 ± 0.5 ^c	4.9 ± 0.2 ^b
STV	2.7 ± 0.2 ^a	3.1 ± 0.2 ^b	5.3 ± 0.2 ^c	7.9 ± 0.3 ^a	8.3 ± 0.3 ^a
T	1.9 ± 0.2 ^c	4.6 ± 0.3 ^a	6.1 ± 0.2 ^b	7.1 ± 0.2 ^b	8.1 ± 0.4 ^a
SA-US	2.1 ± 0.2 ^{bc}	1.0 ± 0.4 ^c	2.1 ± 0.2 ^e	2.8 ± 0.3 ^c	5.2 ± 0.2 ^b
STV-US	1.9 ± 0.2 ^c	4.6 ± 0.3 ^a	6.6 ± 0.2 ^a	7.8 ± 0.3 ^a	8.2 ± 0.3 ^a
T-US	2.5 ± 0.2 ^{ab}	5.2 ± 0.3 ^a	6.8 ± 0.3 ^a	7.3 ± 0.3 ^{ab}	8.3 ± 0.3 ^a

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