

1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17

Influence of olive tree irrigation and the preservation system on the fruit characteristics
of Hojiblanca black ripe olives

P. García, C. Romero, M. Brenes*

Food Biotechnology Department, Instituto de la Grasa-CSIC

Avda. Padre García Tejero 4, 41012-Seville, Spain

* Corresponding author. *E-mail address:* brenes@cica.es (M. Brenes)

Tel: + 34 954690850

18 **Abstract**

19

20 In this study, the effect of olive tree irrigation, the use of salt in preservation liquids and
21 the reuse of sodium hydroxide solutions (lye) on the weight, shriveling, firmness and
22 phenolic content of Hojiblanca processed olives was investigated. A weight loss in
23 fruits of up to 5% during the preservation stage was observed, particularly for olives
24 from irrigated trees and stored in brines. By contrast, a weight gain of up to 7% was
25 achieved during the darkening stage, whose intensity was increased by using fruits from
26 non-irrigated trees and preserved in a salt-free environment as well as fresh lye for the
27 debittering step. Moreover, shriveling particularly appeared in fruits from non-irrigated
28 olive trees, this defect being more intense if lye was reused. Firmness was also affected
29 by the studied variables, and natural rainfed irrigation and the reuse of lye and salt in the
30 preservation solutions gave rise to firmer olives. The content in phenolic compounds of
31 black processed olives was higher in fruits from non-irrigated than irrigated trees, in
32 particular those of hydroxytyrosol, tyrosol and luteolin 7-glucoside. Overall, these
33 results will contribute to the knowledge of table olives processing and the industrial
34 optimization of this sector.

35

36 **Keywords:** olive, weight, shriveling, firmness, phenolic

37

38 **1. Introduction**

39

40 Spain is the major producing country of table olives with ca. 500 million
41 kilograms per year, about 40% of this production being obtained from the Hojiblanca
42 variety, which is an emerging table olive variety worldwide. These fruits are mainly
43 intended for black olives and are mechanically harvested because of their hard texture
44 and low incidence of bruising.

45 The industrial production of black olives involves their harvesting at an early
46 stage of maturation when they have a green/yellow color on the surface, their covering
47 with an acidified brine and oxidation darkening under alkaline conditions (García,
48 Brenes & Garrido, 1991). Considerable research has been carried out on the
49 preservation and darkening process of Hojiblanca black olives (García, Brenes &
50 Garrido, 1991; Brenes, García, Romero & Garrido, 1998; de Castro, García, Romero,
51 Brenes & Garrido, 2007) but changes have been introduced in the process during the
52 last years that make it necessary for industries to know their effects on the
53 characteristics of the final product.

54 The influence of olive tree irrigation on the quality of this product has never
55 been investigated. Indeed, there is limited information about the effect of tree irrigation
56 on the quality of table olives in general (Proietti & Antognozzi, 1996; Marsilio et al.,
57 2006). Besides, changes in olive weight during the preservation and darkening stages of
58 black olive processing is a controversial matter because reliable data are not available.
59 In fact, on many occasions, the processors themselves argue over contradictory data
60 about the weight loss of olives during the preservation stage, which could be related to
61 the presence of salt in the liquids (De Castro et al., 2007). It has been reported that
62 olives of the Verdial variety gained weight during the darkening step (Garrido, Albi &

63 Fernández., 1973) but it was not related to the olive tree irrigation or the storage
64 solution used. Also, the reuse of the sodium hydroxide solutions is another widespread
65 industrial practice that could affect the fruit characteristics (Garrido, 1984).

66 It is well-documented that fruits from irrigated olive trees have lower contents in
67 phenolic compounds (Patumi, d'Andria, Marsilio, Fontanazza, Morelli et al., 2002;
68 Marsilio et al., 2006) than those from non-irrigated trees, as well as lower activity of
69 enzymes involved in the biosynthetic routes of polyphenols (Tovar, Romero, Girona &
70 Motilva, 2002). Irrigation also affects the level of phenolic compounds in olive oils
71 (Stefanoudaki, Williams, Chartzoulakis, & Harwood, 2009) but the effect of water
72 stress on the concentration of phenolic compounds in black olives has never been
73 studied. Water deficit can also affect some table olive quality parameters such as the
74 presence of shriveling on the surface of fruits or the firmness of the final product.

75 The aim of the current study was to provide reliable data about the influence of
76 olive tree irrigation, the presence of salt in the preservation solution and the reuse of lye
77 on some important characteristics of the Hojiblanca fruits such as their weight, presence
78 of shriveling, firmness and content in phenolic compounds.

79

80 **2. Materials and methods**

81 *2.1 Olives*

82 Olives of the Hojiblanca variety were mechanically harvested at a maturity stage
83 (green/yellow color on surface) suitable for processing in October 2009. Fruits were
84 cultivated in Lora de Estepa and Casariche, two small towns in the province of Seville
85 (Spain), under irrigation and non-irrigation conditions. The annual rainfall for 2009 was
86 540 mm, being abundant during the spring and autumn and almost insignificant during
87 the summer. Farmers supplied the olives to two Cooperatives from irrigated or non-

88 irrigated soils over four weeks in October. On arrival, leaves and small branches were
89 removed and the fruits were washed.

90

91 *2.2 Preservation stage*

92 Olives were put in fiberglass underground tanks, which contained about 9500
93 kg of fruits and 5500 L of cover solution. During the four weeks of October, in the two
94 Cooperatives, two different samples of olives from irrigated or non-irrigated trees were
95 put into two tanks (duplicate) and covered with a 3.5 g/100 mL of NaCl and 1.6 g/100
96 mL of acetic acid solution or with just the 1.6 g/100 mL acetic acid solution. A total of
97 128 tanks were used for the experiments. All of the tanks were maintained under
98 aerobic conditions by bubbling air from the bottom of the tank with a column as
99 described elsewhere (De Castro et al., 2007).

100 To study the evolution of the weight of the olives during preservation, 4.0 kg of
101 fruits were put into a plastic net and introduced at 1 m of depth into each tank with a
102 plastic string. Periodically, at 1, 4, 7 and 10 months from preservation the plastic nets
103 were removed from the tanks and the olives were weighed.

104

105 *2.3 Darkening stage*

106 The olives stored for 7-8 months were darkened as black ripe olives in 12 PVC
107 cylindrical containers with conical bases (García, Brenes & Garrido, 1991). Three olive
108 samples from each non-irrigated tree and brine storage, non-irrigated tree and salt-free
109 storage, irrigated tree and brine storage, and irrigated tree and salt-free storage
110 treatments were darkened. The process consisted of placing 1.5 kg of fruits in 1.5 L of
111 0.75 mol/L of NaOH solution (lye) for 4-5 h, sufficient time for the lye to reach the pit.
112 The olives were then covered with tap water and air was bubbled through the mixture

113 for 24 h. After draining, the olives were put in a new washing solution (tap water:
114 preservation olive solution, 1:1), and air was bubbled for 24 h (Brenes et al., 1998).
115 Finally, the liquid was poured off and the fruits were covered with a 0.1 g/100 mL of
116 ferrous gluconate solution and aerated for another 24 h. Before packing, the weight of
117 the olives was checked.

118 A weighed amount of whole (ca. 175 g) and pitted (ca. 145 g) fruits were bottled in
119 cylindrical A314 jars (Juvasa, Seville, Spain) with 145 and 175 mL of a cover solution
120 respectively, which had 3 g/100 mL of NaCl and 0.025 g/100 mL of ferrous gluconate.
121 Calcium chloride (0.35 g/100 mL) was also added in the cover solution of half of the
122 jars. All of them were sterilized at 121°C for 15 min in a computer-controlled Steriflow
123 retort (Madinex, Barcelona, Spain). One month from packing, they were opened and the
124 olives were weighted. Firmness and shriveling were tested.

125 Olive samples from treatments reported above were also processed with reused
126 lye supplied by the Agrosevilla SCA factory instead of fresh lye. The concentration of
127 this reused lye was adjusted to 0.75 mol/L of NaOH.

128

129 *2.4 Changes in volume of olives during lye treatment*

130 One liter of tap water was put into a 2 L graduated cylinder, and 1 kg of olives
131 was added. The increase in volume of the mixture was recorded. Then, fruits were
132 treated with a 0.75 mol/L of NaOH solution until the alkaline solution reached the pit,
133 which was monitored by adding a drop of phenolphthaleine ethanolic solution on the
134 pulp of olives. Subsequently, the fruits were weighed and put into the graduated
135 cylinder containing 1 L of water. The difference between the volume of olives before
136 and after the lye treatment was recorded.

137

138 *2.5 Quality analysis of olives*

139

140 Firmness of olives was measured using a Kramer shear compression cell coupled
141 to an Instron Universal Testing Machine Model 1001 (Canton, USA). The crosshead
142 speed was 200 mm/min. Firmness was the mean of 10 replicate measurements, each of
143 which was performed on 3 pitted olives, and expressed as N/100 g pitted olives.
144 Analyses were made one month after packing.

145 The presence of olives with wrinkles that affected their appearance was tested by
146 three table olive experts on 100 olive fruits one month after packing. This was
147 expressed as % of shriveled fruits.

148

149 *2.6 Polyphenols analysis*

150

151 Pitted fruits were crushed with an Ultraturrax homogenizer, and 3 g of the paste
152 were mixed with 18 ml of dimethyl sulfoxide (DMSO) (Romero-Segura, Sanz, & Pérez,
153 2009). After 1 min of agitation by vortex, the mixture was centrifugated at 6000 g for 5
154 min, and the supernatant was filtered through a 0.22 µm pore size nylon filter. An
155 aliquot of 250 µL was mixed with 250 µL of internal standard (0.2 mmol/L of syringic
156 acid in DMSO) and 500 µL of DMSO. Finally, 20 µL of the mixture were injected into
157 the chromatograph. A Spherisorb ODS-2 (5 µm, 250 x 4.6 mm, Waters Inc.) column
158 was used. The HPLC system consisted of a Waters 2695 Alliance (Waters Inc.,
159 Mildford, MA, USA) with a pump, column heater and autosampler included, the
160 detection being performed with a Waters 996 diode array detector at 280 nm. Separation
161 was achieved using an elution gradient with an initial composition of 90% water (pH
162 adjusted to 2.3 with phosphoric acid) and 10% methanol. The concentration of the latter

163 solvent was increased to 30% over 10 min and maintained for 20 min. Subsequently, the
164 methanol % was raised to 40% over 10 min, maintained for 5 min and then increased to
165 50%. Finally, the methanol % was increased to 60%, 70% and 100% in 5 min intervals.
166 The flow rate of 1 mL/min and a temperature of 35°C were used (Medina et al., 2007).

167

168 2. 7 Statistics

169 Statistical comparisons of the mean values for each experiment were performed by
170 one-way analysis of variance (ANOVA), followed by the Duncan's multiple range test
171 ($p < 0.05$) using Statistica software version 6.0 (Stat-Soft, 2001).

172

173

174 3. Results and discussion

175

176 There was a continuous weight loss of the fruits with time in all tanks (Fig. 1), although
177 significant differences among the treatments were detected. Olives from non-irrigated
178 trees lost weight to a lower extent than fruits from irrigated trees, and salt in the
179 preservation solution gave rise to faster weight loss. After 10 months of preservation,
180 the lowest (3.5%) and the highest (5.1%) weight loss was obtained for olives from non-
181 irrigated trees and preserved in a salt-free solution, and olives from irrigated trees and
182 preserved in acidified brine, respectively. Moreover, the time of harvesting did not show
183 a statistically significant effect ($p < 0.05$) on the weight loss (data no shown) but fruits
184 harvested at the end of October tend to have higher weight loss than those picked during
185 the first week of the month. It must be said that these results were obtained for the
186 Hojiblanca variety and the intensity of the weight loss for other olive varieties could be
187 different. However, we have collected sporadic data for years and the tendency was

188 quite similar for all the varieties. The diffusion of substances from the olives to the
189 preservation solution seems to explain the weight loss. Also, this phenomenon was
190 more intense in olives submerged in brine because of the osmotic action of the salt.

191 In contrast to the weight loss found during the preservation stage, there was a significant
192 weight gain in the darkening process, especially during the NaOH treatment (Fig. 2).

193 Overall, olives gained about 5-7% of weight, a percentage higher than the weight loss
194 which occurred during the preservation stage, although this gain depended on several
195 factors. The use of fresh lye gave rise to a higher weight gain in olives than reused lye
196 ($p < 0.05$). Despite these results, factories must continue reusing the lye in order to
197 reduce the environmental impact of table olive wastewaters. Significant weight gain
198 differences were also found between olives from irrigated and non-irrigated trees,
199 having gained the former a higher percentage of weight than the later. It must be
200 remembered that olives from irrigated trees lost a higher percentage of weight than non-
201 irrigated during the preservation stage. Moreover, the presence of salt in the
202 preservation solution provoked a lower weight gain during darkening, which is another
203 argument in favor of eliminating NaCl from the preservation solutions of black olives as
204 well as its contribution to the mineral contamination of the table olive wastewaters.

205 There are only few previous data about the effect of the darkening process on the weight
206 of olives (Garrido, Albi & Fernández, 1973) but no explanation for this phenomenon
207 was offered. A very simple experiment was done to clarify this issue. One kilogram of
208 olives was put in contact with an NaOH solution (0.75 mol/L) for 5 hours and a
209 significant increase in weight (11.3%), as well as in volume, was observed.

210 Additionally, fruits increased their volume from 0.6 L to 0.7 L (Fig. 3). It seems that the
211 weight gain of olives during the darkening stage, in particular during the alkaline
212 treatment, was due to the increase in volume of the fruits and, consequently, in weight.

213 It has been reported that the lye treatment affects both the skin and mesocarp textural
214 characteristics (Georget, Smith, Waldron, & Rejano, 2003), it dissolves the epicuticular
215 waxy coating and breaks the intercellular pectic material in the middle lamella causing
216 the softening of tissues (Marsilio, Lanza & De Angelis, 1996). Moreover, changes in the
217 intercellular volume of olives have also been reported as a consequence of gas diffusion
218 (oxygen, nitrogen and carbon dioxide) during their preservation stage (Romero, Brenes,
219 García & Garrido, 1996), and even the growth of lactic acid bacteria could provoke
220 structural modification in tissues (Servili et al., 2008). However, an increase in cell
221 volume due to the lye treatment has never been reported. Thus, it can be speculated that
222 the volume gain of olives during this alkaline treatment, and therefore weight gain,
223 could be due to cell separation because of pectic material solubilization of the middle
224 lamella.

225 Olives also suffered small weight changes after packing and sterilization. Whole fruits
226 lost weight (ca. 1.1%), whereas pitted olives gained weight (ca. 0.6%). Neither the
227 preservation method nor the irrigation regime had a significant effect on the weight
228 changes in the olives after packing (data not shown).

229 The presence of wrinkles on the surface of olives sometimes becomes a severe
230 commercial defect that makes them unacceptable to consumers. This phenomenon is
231 rare for Manzanilla olives but common for Hojiblanca fruits. Many variables can be
232 involved in this damage, such as the preservation method. If anaerobic conditions are
233 maintained, a very high concentration of carbon dioxide will be accumulated in the
234 solutions and olives. This gas is released from the fruits when they are taken out of the
235 fermenters and small wrinkles can appear on the olive surface. By contrast, the
236 maintenance of aerobic conditions prevents this damage because carbon dioxide is

237 constantly purged (Romero et al., 1996). In our industrial trials, aerobic conditions were
238 used.

239 Other variables can also affect the presence of shriveling on fruits. Fig. 4 shows the
240 influence of tree irrigation and type of lye on the percentage of shriveling on whole and
241 pitted packed olives. First, we must say that the statistical analysis ruled out the
242 presence of salt in the preservation solution as a key factor for shriveling on olives (data
243 not shown). By contrast, the stress water of the olive trees exerted a significant effect on
244 this alteration, in particular when lye was reused. Also, the effect was more severe on
245 whole than pitted olives. We do not have an explanation for this phenomenon although
246 Patumi et al. (2002) indicated that olive trees under water stress conditions produced
247 fruits with a higher cuticular thickness to prevent the loss of water, and it could
248 influence the presence of shriveling when olives are processed.

249 Results from our experiments also disclosed that firmness was another quality
250 characteristic of olives influenced by the tree irrigation regime, the reuse of lye and the
251 presence of salt in the preservation solution (Fig. 5). Fruits cultivated in irrigated trees
252 had significantly lower firmness than those from rainfed trees, which is in accordance
253 with previous works (Proietti & Antognozzi, 1996; Marsilio et al., 2006). In contrast to
254 the results obtained by Garrido (1984), firmer olives were obtained when reusing the
255 lye. Moreover, it was confirmed that the presence of a low level of salt in the
256 preservation solution gave rise to harder olives (De Castro et al., 2007). Several studies
257 have shown the positive effect of sodium ions on olive texture (García, Brenes &
258 Garrido, 1994; De Castro et al., 2007) and other fermented vegetables (Fleming,
259 McFeeters & Thompson, 1987). In the case of Hojiblanca fruits, firmness is not
260 currently a key quality parameter because of their hard texture (Georget, Smith &
261 Waldron, 2001), except when they are preserved in a very highly acidified medium free

262 of salt. In order to solve this softening problem, calcium ions were added to the packing
263 solutions of olives preserved in a salt-free environment, and they showed a similar
264 texture to those stored in acidified brine without calcium added during packing (Fig. 6),
265 thereby confirming the great influence that calcium ions can exert on the firmness of
266 table olives (Romero, García, Brenes & Garrido, 1995; Tassou et al., 2007; De Castro et
267 al., 2007).

268 Olive polyphenols play an important role in table olive characteristics since they
269 contribute to their color, flavor and texture. Besides, these substances are powerful
270 antioxidants and exert many beneficial effects on human health. The effects of tree
271 irrigation, salt in the preservation solution and the reuse of lye on the phenolic content
272 of packed black olives were studied. After the statistical analysis of the data, the
273 irrigation regime only showed a significant effect on the concentration of these
274 substances in olives (Fig. 7). The total content of phenolic compounds in olives from
275 non-irrigated and irrigated trees was 602 and 501 mg/kg, respectively. Differences were
276 found among all the individual compounds although they were only significant for
277 hydroxytyrosol, salidroside, tyrosol and luteolin 7-glucoside. The effect of water stress
278 on the phenolic content of olive fruits (Patumi et al., 2002; Tovar et al, 2002), olive oil
279 (Stefanoudaki et al, 2009) and naturally green processed olives (Marsilio et al., 2006) is
280 well-documented, the higher the irrigation regime the lower the content in phenolic
281 compounds. With regard to black oxidized olives, we observed an increase of about
282 20% in phenolic concentration between fruits from irrigated and non-irrigated olive
283 trees. It is not a very high difference but it must be noted that olives suffered many
284 water changes during processing.

285

286 **4. Conclusions**

287 The weight and volume of black olives during processing is affected by the type of olive
288 tree irrigation, the use of salt in the preservation solution and the reuse of lye. Fruits lost
289 and gained weight during the preservation and darkening steps respectively. These
290 changes are dependent on the above mentioned variables. In addition, the non-irrigation
291 of olive trees and the reuse of lye influenced the presence of shriveling on the surface of
292 the olives to a great extent. All these data will contribute to the optimization of the black
293 ripe olive processing, and to explain some changes that occur during this elaboration
294 process at industrial scale.

295

296

297 **Acknowledgements**

298 This work was supported by the Projects AGL-2009-07512 and AGL-2010-15494.

299 Thanks to Agrosvilla SCA for collaboration.

300

301 **References**

302

303 Brenes, M., García, P., Romero, C., & Garrido, A. (1998). Ripe olives storage liquids
304 reuse during the oxidation process. *Journal of Food Science*, *63*, 117-121.

305 De Castro, A., García, P., Romero, C., Brenes, M., & Garrido, A. (2007). Industrial
306 implementation of black ripe olive storage under acid conditions. *Journal of Food*
307 *Engineering*, *80*, 1206-1212.

308 Fleming, H. P., McFeeters, R. F., & Thompson, R. L. (1987). Effects of sodium
309 chloride concentration on firmness retention of cucumbers fermented and stored
310 with calcium chloride. *Journal of Food Science*, *52*, 653-657.

311 Garrido, A., Albi, M. A., & Fernández, M. J. (1973). Black olives by alkaline oxidation.
312 II. Iron determination and development of several factors during processing. *Grasas*
313 *y Aceites*, *24*, 287-292.

314 Garrido, A. (1984). Study of wastewaters from ripe olive processing (and II). Effect of
315 reusing lye or aeration solution on colour, texture and canning brines. *Grasas y*
316 *Aceites*, *35*, 165-171.

317 García, P., Brenes, & M., Garrido, A. (1991). Effect of oxygen and temperature on the
318 oxidation rate during the darkening step of ripe olives processing. *Journal of Food*
319 *Engineering*, *13*, 259-271.

320 García, P., Brenes, M., & Garrido, A. (1994). Effects of pH and salts on the firmness of
321 canned ripe olives. *Sciences des Aliments*, *14*, 159-172.

322 Georget, D. M. R., Smith, A. C., & Waldron, K. W. (2001). Effect of ripening on the
323 mechanical properties of Portuguese and Spanish varieties of olive (*Olea europaea*
324 L.). *Journal of the Science of Food and Agriculture*, *81*, 448-454.

- 325 Georget, D. M. R., Smith A. C., Waldron, K. W., & Rejano, L. (2003). Effect of
326 “Californian” process on the texture of Hojiblanca olive (*Olea europaea* L.)
327 harvested at different ripening stages. *Journal of the Science of Food and*
328 *Agriculture*, 83, 574-579.
- 329 Marsilio, V., Lanza, B., & De Angelis, M. (1996). Olive cell wall components: physical
330 and biochemical changes during processing. *Journal of the Science of Food and*
331 *Agriculture*, 70, 35-43.
- 332 Marsilio, V., d’Andria, R., Lanza, B., Russi, F., Iannucci, E., Lavini, A., & Morelli, G.
333 (2006). Effect of irrigation and lactic acid bacteria inoculants on the phenolic
334 fraction, fermentation and sensory characteristics of olive (*Olea europaea* L. cv.
335 Ascolana tenera) fruits. *Journal of the Science of Food and Agriculture*, 86, 1005-
336 1013.
- 337 Medina, E., Brenes, M., Romero, C., García, A., & de Castro, A. (2007). Main
338 antimicrobial compounds in table olives. *Journal of Agricultural and Food*
339 *Chemistry*, 55, 9817-9823.
- 340 Patumi, M., d’Andria, R., Marsilio, V., Fontanazza, G., Morelli, G., & Lanza, B. (2002).
341 Olive and olive oil quality alter intensive monocone olive growing (*Olea europaea*
342 L., cv. Kalmata) in different irrigation regimes. *Food Chemistry*, 77, 27-34.
- 343 Proietti, P., & Antognozzi, E. (1996). Effect of irrigation on fruit quality of table olives
344 (*Olea europaea*) cultivar “Ascolana tenera”. *New Zealand Journal of Crop and*
345 *Horticultural Science*, 24, 175-181.
- 346 Romero, C., García, P., Brenes, M., & Garrido, A. (1995). Colour and texture changes
347 during sterilization of packed ripe olives. *International Journal of Food Science and*
348 *Technology*, 30, 31-36.

- 349 Romero, C., Brenes, M., García, P., & Garrido, A. (1996). *Respiration of olives stored*
350 *in sterile water. Journal of Horticultural Science, 71, 739-745.*
- 351 Romero-Segura, C., Sanz, C., & Pérez, A. G. (2009). Purification and characterization
352 of olive fruit β -glucosidase involved in the biosynthesis of olive oil phenolics.
353 *Journal of Agricultural and Food Chemistry, 57, 7983-7988.*
- 354 Servili, M., Minnocci, A., Veneziani, G., Taticchi, A., Urbani, S., Esposito, S.,
355 Sebastiani, L., Valmorri, S. & Corsetti, A. (2008). Compositional and tissue
356 modifications induced by the natural fermentation process in table olives. *Journal of*
357 *Agricultural and Food Chemistry, 56, 6389-6396.*
- 358 Stefanoudaki, E., Williams, M., Chartzoulakis, K., & Harwood, J. (2009). Effect of
359 irrigation on quality attributes of olive oil. *Journal of Agricultural and Food*
360 *Chemistry, 57, 7048-7055.*
- 361 Tassou, C. C.; Katsaboxakis, C. Z., Georget, D. M. R., Parker, M. L., Waldron, K. W.,
362 Smith, A. C., & Panagou, E. Z. (2007). Effect of calcium chloride on mechanical
363 properties and microbial characteristics of cv. *Conservolea* naturally black olives
364 fermented at different sodium chloride levels. *Journal of the Science of Food and*
365 *Agriculture, 87, 1123-1131.*
- 366 Tovar, M. J., Romero, M. P., Girona, J., & Motilva, M. J. (2002). L-Phenylalanine
367 ammonia-lyase activity and concentration of phenolics in developing olive (*Olea*
368 *europaea* L. cv. Arbequina) fruit grown under different irrigation regimes. *Journal*
369 *of the Science of Food and Agriculture, 82, 892-898.*
- 370
- 371
- 372
- 373

374

375 Figure legends

376

377 **Fig. 1.** The effects of tree irrigation and the presence of salt in the preservation solution
378 on the weight loss of Hojiblanca olives during their storage before the darkening stage:
379 (◆) non-irrigated trees and brine storage, (●) non-irrigated trees and salt-free storage,
380 (▲) irrigated trees and brine storage, (■) irrigated trees and salt-free storage. The error
381 bars represent the standard errors of 32 replicates.

382

383 **Fig. 2.** The effects of the reuse of lye, tree irrigation, and salt in the preservation
384 solution on the weight gain of Hojiblanca olives during the darkening process. The error
385 bars represent the standard errors of 12 replicates. Different letters mean significant
386 differences according to a Duncan's multiple range test ($P<0.05$).

387

388 **Fig. 3.** The effect of the alkaline treatment on the weight and volume of Hojiblanca
389 olives.

390

391 **Fig. 4.** The effects of tree irrigation and the reuse of lye on the shriveling defect of
392 Hojiblanca black olives: ■ irrigated trees and fresh lye, □ non-irrigated trees and fresh
393 lye, ▨ irrigated trees and reused lye, ▩ non-irrigated trees and reused lye. Olive
394 quality was tested after one month from olive packing. The error bars represent the
395 standard errors of 6 replicates. Different letters mean significant differences according
396 to a Duncan's multiple range test ($P<0.05$).

397

398 **Fig. 5.** The effects of the reuse of lye, tree irrigation and the presence of salt in the
399 preservation solution on the firmness of Hojiblanca black olives. Measurements were
400 made after one month from olive packing. The error bars represent the standard errors
401 of 12 replicates. Different letters mean significant differences according to a Duncan's
402 multiple range test ($P<0.05$).

403

404 **Fig. 6.** The effect of calcium addition in the packing solutions on the firmness of
405 Hojiblanca black olives one month after sterilization. Analyses were made after one
406 month from olive packing. The error bars represent the standard errors of 6 replicates.
407 Different letters mean significant differences according to a Duncan's multiple range
408 test ($P<0.05$).

409

410 **Fig. 7.** The effect of tree irrigation on the content of phenolic compounds in the
411 Hojiblanca black olives. Analyses were made after one month from olive packing. The
412 error bars represent the standard errors in triplicate. Different letters mean significant
413 differences according to a Duncan's multiple range test ($P<0.05$).

414

Figure 1

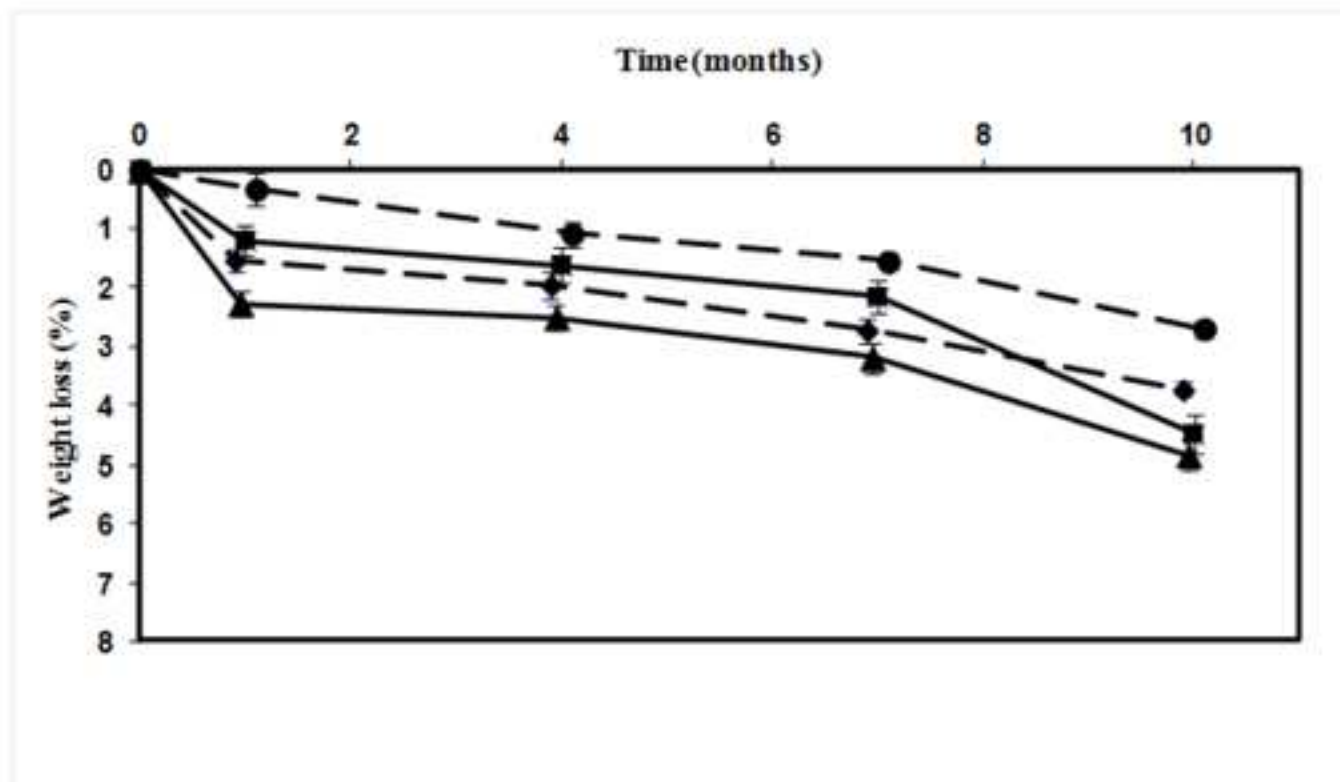


Figure 2

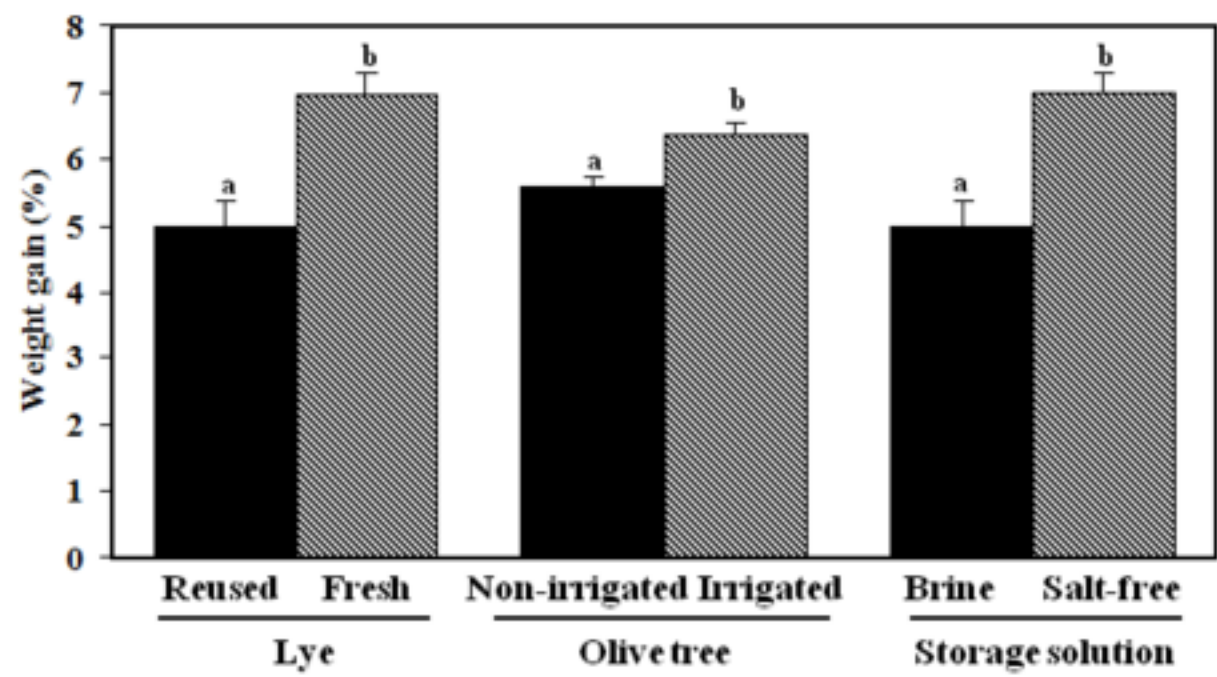


Figure 3

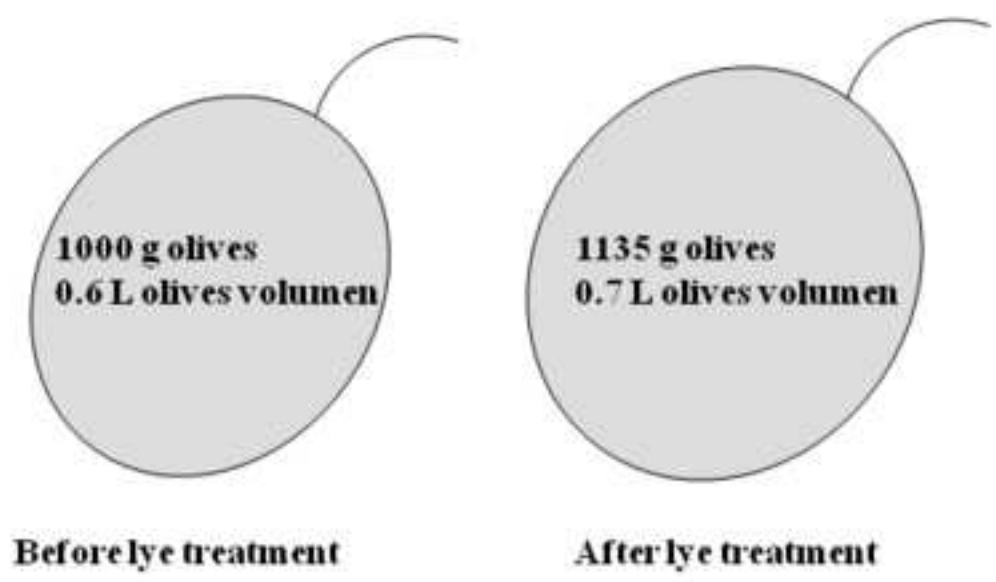


Figure 5

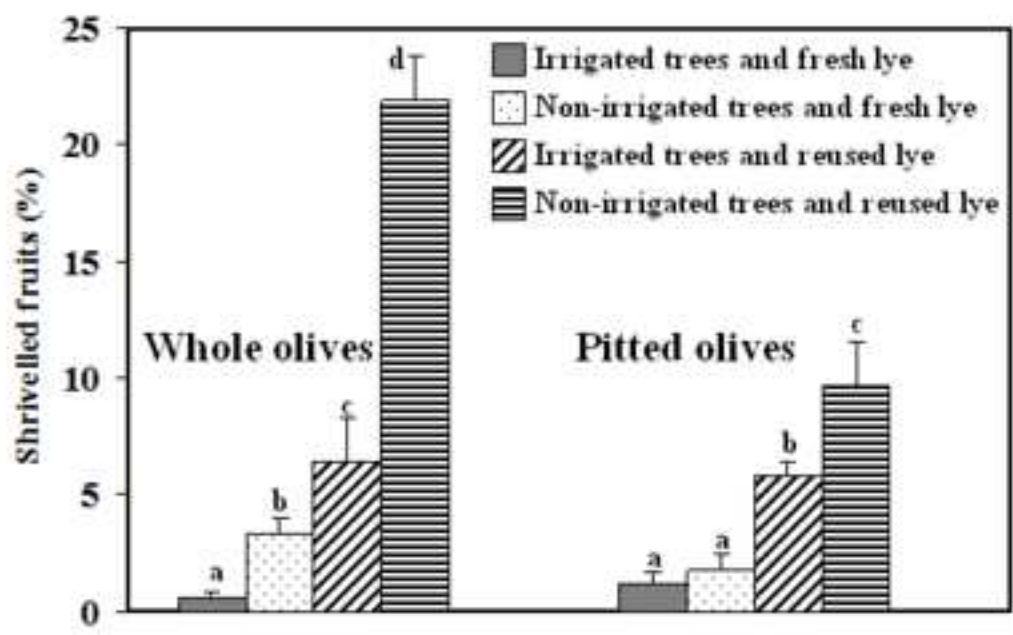


Figure 6

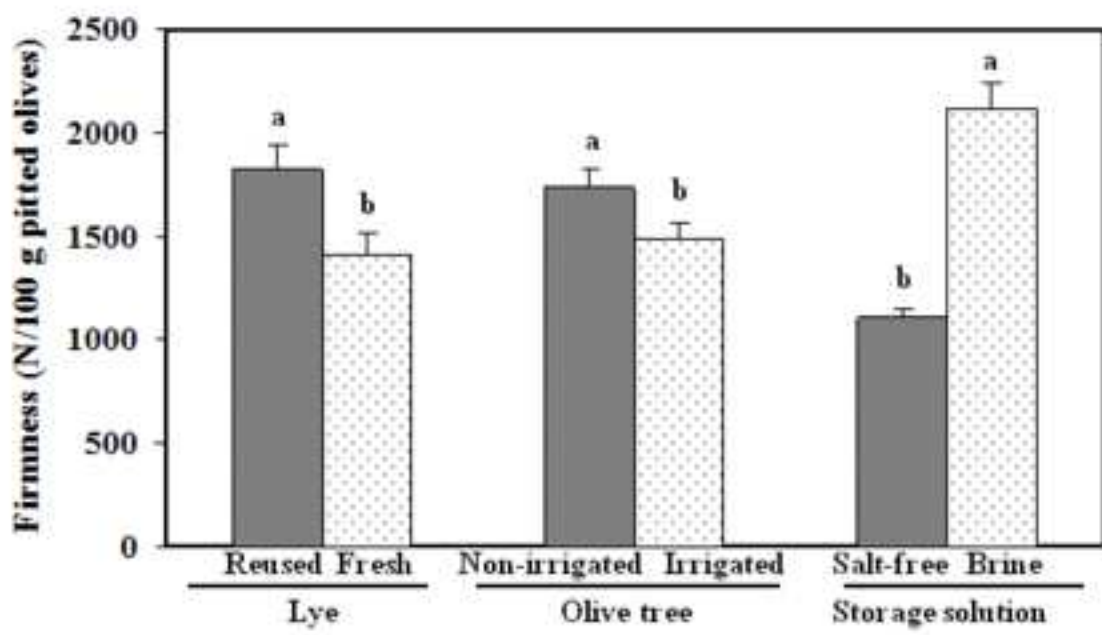


Figure 7

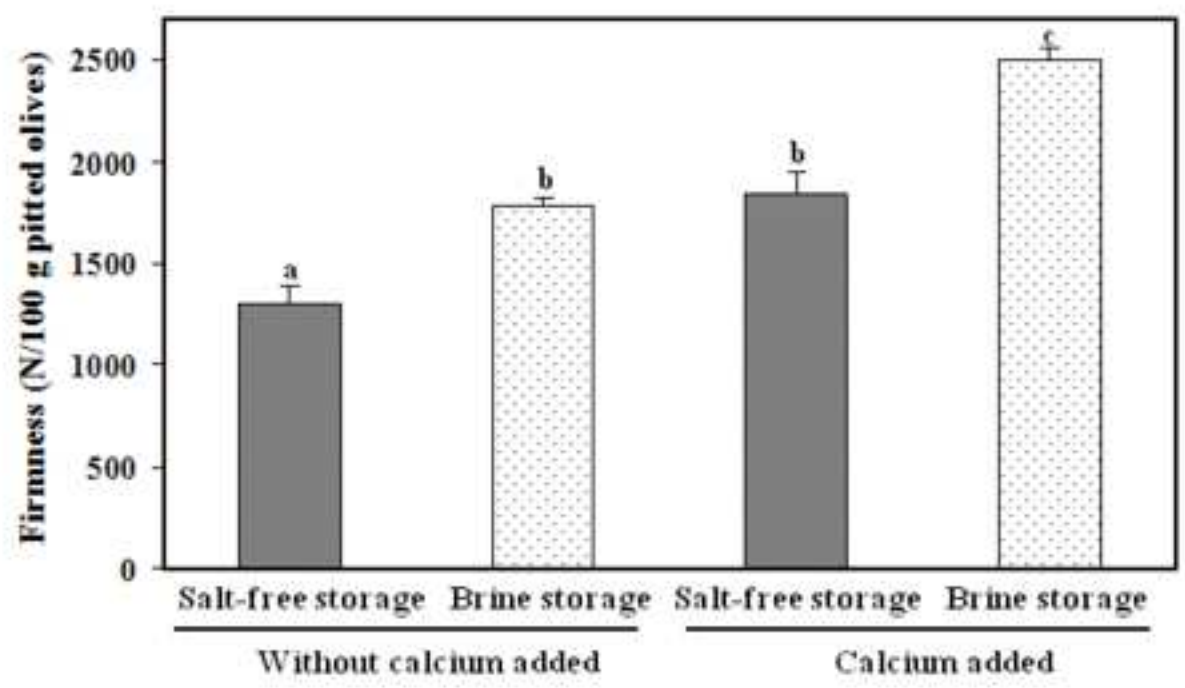


Figure 8

