

1 **Influence of peach–almond hybrids and plum-based rootstocks on mineral**
2 **nutrition and yield characteristics of ‘Big Top’ nectarine in replant and heavy-**
3 **calcareous soil conditions**

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5 **Lucía Mestre¹, Gemma Reig², Jesús A. Betrán¹, Jorge Pinochet³ and María**
6 **Ángeles Moreno^{2*}**

7 ¹Dpto. de Suelos y Aguas de Riego, Laboratorio Agroambiental. Gobierno de Aragón,
8 Apdo. 727, 50080 Zaragoza, Spain

9 ²Dpto. de Pomología, Estación Experimental de Aula Dei (CSIC), Apdo. 13034, 50080
10 Zaragoza, Spain

11 ³Agromillora Iberia S.L., C/El Rebato s/n, 08739 Subirats, Barcelona, Spain

12
13 *Corresponding author. Tel: +34 976 71 61 36. Fax: +34 976 71 61 45; E-mail address:
14 mmoreno@ead.csic.es

15
16 **Abstract**

17 The agronomic performance and leaf mineral nutrition for ‘Big Top’ nectarine budded
18 onto twelve *Prunus* rootstocks were evaluated. Seven *Prunus amygdalus* × *Prunus*
19 *persica* hybrids (Adafuel, Adarcias, Felinem, Garnem, Monegro, GF 677, and Mayor),
20 two *Prunus davidiana* × *P. persica* hybrids (Barrier, Cadaman), a *Prunus insititia* plum
21 (Adesoto), a *Prunus domestica* plum (Tetra), and another selection considered to be an
22 hybrid of *Prunus cerasifera* × *P. amygdalus* parentage (Replantpac). Rootstocks were
23 budded during the summer of 1999, and trees were established in a replant site in
24 March 2001. The trial was located in the Ebro Valley (Northeastern, Spain) on a heavy-
25 textured and calcareous soil typical of the Mediterranean area which supported a
26 previous peach orchard until 2000. At the thirteenth year after budding, growing
27 conditions generated varying levels of tree mortality, the highest with peach-almond
28 hybrids: Adafuel, Garnem and Monegro. In contrast, all Replantpac trees survived well
29 and the mortality rate was low on the other rootstocks. Adesoto, Tetra, and Adarcias
30 proved to be the most dwarfing rootstocks, while Cadaman and Replantpac were the
31 most invigorating and generated greater cumulative yields. However, the highest yield
32 efficiency was recorded on GF 677, although it did not differ significantly from other
33 peach-almond (Adarcias, Felinem) and plum (Adesoto, Tetra) rootstocks. The highest
34 fruit weight was observed on Barrier and the lowest on Felinem and Mayor, but they

35 did not differ significantly from the rest of rootstocks. Leaf mineral analysis of trees
36 showed all rootstocks induced N and Fe deficiency and P optimum value according to
37 reference values. Nevertheless, the tendency of plum Adesoto to induce higher Fe leaf
38 concentration could indicate higher tolerance to iron-chlorosis in calcareous soils. The
39 most invigorating rootstock Replantpac seems to induce higher SPAD values and
40 adequate K, Mg and Mn values according to reference values. Tetra induced the best
41 balanced nutritional values (Σ DOP), especially when compared with Barrier and
42 Cadaman, although it did not differ significantly from GF 677 and Mayor.

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44 **Keywords:** Interspecific hybrids, chlorosis, vigour, foliar mineral analysis

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46 **1.Introduction**

47 Peach [*Prunus persica* (L.) Batsch] is the most important temperate and deciduous fruit
48 tree grown in the world, after apples. Spain is the third leading peach producer in the
49 world, only surpassed by China and Italy, and the second larger producer in the EU,
50 after Italy (FAOSTAT, 2014). The main peach producing area is the Ebro Valley, which
51 includes regions of Aragon and Catalonia, and it accounts for 63% of the total Spanish
52 peach production (MAGRAMA, 2014).

53 Different studies with *Prunus* spp. (Font i Forcada et al., 2012, 2014; Giorgi et al.,
54 2005; Jiménez et al., 2007, 2011; Loreti and Massai, 2006; Moreno et al., 1994, 2001;
55 Remorini et al., 2008; Zarrouk et al., 2005) revealed that the rootstock influences the
56 agronomic performance (tree vigour, yield efficiency, water relations, leaf gas
57 exchange, mineral nutrients uptake, plant size, bloom and harvest dates, and fruit bud
58 survival). The rootstock choice represents one of the most important considerations for
59 a productive peach orchard, particularly in a replant situation (Jiménez et al., 2011;
60 Orazem et al., 2011; Reighard et al., 1997). The use of rootstocks is mainly directed to
61 overcome soil and disease problems to which scions have limited or no resistance.
62 Peach-almond hybrids (*Prunus amygdalus* × *P. persica*) are largely used as rootstocks
63 for peach trees in the Mediterranean countries. They are tolerant to lime induced iron-
64 chlorosis and alkaline soil conditions, and they are graft-compatible with peach and
65 almond cultivars (Moreno and Cambra, 1994; Moreno et al., 1994; Zarrouk et al.,
66 2005). They are also vigorous and appropriate for use in poor dry soils (Cambra, 1990)
67 and in fruit tree replanting situations (Jiménez et al., 2011; Orazem et al., 2011). In
68 recent years, new selections of peach-almond hybrids have also been developed with

69 resistance to biotic stresses, such as root-knot nematodes (*Meloidogyne* spp.) (Felipe,
70 2009; Pinochet, 1997, 2009), and tolerance to replant conditions (Jiménez et al., 2011).
71 Similarly, several plum rootstocks used for different stone fruit species have also been
72 released. They adapt well to highly calcareous and heavy-textured soils, being tolerant
73 to root asphyxia and Fe chlorosis and resistant to root-knot nematodes (Moreno et al.,
74 1995a, 1995b).

75 The present research was carried out over thirteen years of study with ‘Big Top’
76 nectarine cultivar budded onto different peach-based diploid rootstocks (almond ×
77 peach, peach × *Prunus davidiana*), hexaploid plums and an almond-myrobalan diploid
78 hybrid of different vigour and grown on a heavy and calcareous soil typical of the
79 Mediterranean area, in a replant site. The objective was to evaluate the performance of
80 the rootstocks in these conditions, through tree survival, leaf mineral status, vegetative
81 growth, and yield characteristics.

82 **2. Materials and methods**

83 *2.1. Plant material and trial characteristics*

84 Twelve *Prunus* rootstocks, including seven *Prunus amygdalus* × *Prunus persica*
85 hybrids: Adafuel, Adarcias, Felinem, Garnem, Monegro, GF 677 and Mayor; two
86 *Prunus davidiana* × *P. persica* hybrids: Barrier and Cadaman; one *Prunus insititia*
87 plum: Adesoto; one *Prunus domestica* plum: Tetra; and one *Prunus cerasifera* × *P.*
88 *amygdalus* hybrid: Replantpac, were evaluated since the third (2003) to the thirteenth
89 (2013) year after planting at the Experimental Station of Aula Dei-CSIC (Zaragoza,
90 Spain) (Table 1). Adafuel (Cambra, 1990), Adarcias (Moreno and Cambra, 1994) and
91 Mayor (Cos et al., 2004) were selected due to their tolerance to iron chlorosis. The
92 hexaploid plum Adesoto was selected due to its resistance to root-knot nematodes and
93 good graft-compatibility with peach (Moreno et al., 1995a). Replantpac (Rootpac[®] R)
94 shows resistance to root-knot nematodes and exhibits a high tolerance to root asphyxia
95 caused by waterlogging (Pinochet, 2010). Felinem, Garnem and Monegro were selected
96 due to their tolerance to iron chlorosis and resistance to root-knot nematodes (Fernández
97 et al., 1994; Felipe, 2009). GF 677 is the most commonly used peach × almond hybrid
98 rootstock in Mediterranean countries due to its tolerance to lime induced iron-chlorosis
99 and good agronomical performance (Moreno et al., 1994).

100 These rootstocks were budded with ‘Big Top’ nectarine cultivar during the summer of
101 1999, and trees were established in an experimental plot on March 2001. ‘Big Top’
102 nectarine is an American cultivar (Zaiger breeding program, USA) highly valued and

103 widespread in the European Union in the last decade (Iglesias, 2010). This nectarine is a
104 mid-season reference cultivar, known for its early coloration resulting in highly colored
105 fruit, sweet taste and optimum fruit size (Della Strada and Fideghelli, 2003; Bellini et
106 al., 2004).

107 The trial was located in the Ebro Valley (North-Eastern of Spain), on a heavy and
108 calcareous soil, with 28% total calcium carbonate, 8% active lime, water pH 8.4, and a
109 clay-loam texture. Trial was established on a non-fumigated replant site, one year after
110 uprooting an 8-year-old peach ('Summergrand' nectarine cv.) orchard that was budded
111 on plums (*P. insititia*, *P. domestica*) and peach-almond rootstocks. The experiment was
112 established in a randomized block design with five single-tree replications for each
113 scion-rootstock combination. Guard rows were used to preclude edge effects. Trees
114 were planted at 5.5 m × 5.5 m and trained to a low density open-vase system. Cultural
115 management practices, such as fertilization, winter pruning, and spring thinning, were
116 conducted as in a commercial orchard. Open vase trees were pruned to strengthen
117 existing scaffold branches and eliminate vigorous shoots, inside and outside the vase,
118 that would compete with selected scaffolds or shade fruiting wood. Moderate-sized
119 fruiting wood (0.3-0.6 m long) was selected. All trees were hand-thinned at 45-50 days
120 after full bloom (DAFB) leaving approximately 20 cm between fruits. The plot was
121 level-basin irrigated every 12 days during the summer.

122 *2.2. Tree survival and suckering*

123 Tree health and survival were monitored throughout the trial. Dead trees were recorded
124 each year at time when growth measurements were taken. The incidence of rootstock
125 suckering (root and collar suckers) was also recorded during this study.

126 *2.3. Growth measurements and yield characteristics*

127 For all the cropping years, starting in 2003, trunk girth, yield and number of fruits per
128 tree were recorded. Trunk girth was measured each dormant season at 20 cm above the
129 graft union, and the trunk cross-sectional area (TCSA) was then calculated. At harvest,
130 all fruits from each tree were counted and weighted to determine total yield per tree
131 (kg/tree). Fruit weight (FW) was calculated considering the total number of fruits and
132 total yield per tree. Average fruit weight (AFW) from 2009 to 2013 was also calculated.
133 Cumulative yield (CY) per tree and yield efficiency (YE) of each scion-stock
134 combination were computed from the harvest data. YE was calculated as the ratio
135 between the cumulative yields in kilograms per tree (from 2003 to 2013) per final
136 TCSA (cm²) determined in the winter of 2013-2014.

137 *2.4. Chlorophyll analysis*

138 The chlorophyll (Chl) concentration per unit leaf area was estimated in the field, using a
139 SPAD 502 meter (Minolta Co., Osaka, Japan). After calibration, SPAD measurements
140 were converted into Chl concentration per unit of leaf area (nmol Chl cm⁻²). Thirty
141 leaves per tree, selected from the middle of bearing shoots located all around the crown,
142 were measured with the SPAD to obtain an average leaf Chl concentration
143 representative of the leaves belonging to the outer part of the tree canopy.
144 Measurements were carried out 120 days after full bloom (DAFB) in 2012, as
145 performed during the previous years (Pinochet, 2010).

146 *2.5. Mineral analysis*

147 Leaf mineral element concentrations were determined in 2012, i.e. in year 12 after
148 budding, for 'Big Top' trees with no asphyxia symptoms and/or associated diseases.
149 Leaf sampling was carried out at 120 DAFB. Leaf samples (40 leaves per tree) were
150 collected from shoots around the crown of the trees. The mineral element composition
151 of the dried tissue was determined using the methods of C.I.I (1969) and C.I.I et al.
152 (1975), as previously reported by Jiménez et al. (2007). Total N was determined by
153 Kjeldahl analysis (Gerhardt Vapodest); P was analyzed spectrophotometrically by the
154 phospho-vanadate colorimetric method (Hewlett-Packard 8452A); K, Ca, Mg and Na by
155 atomic emission spectroscopy (ICP, Horiba-Jobin Yvon, Activa-M); and Fe, Mn, Cu
156 and Zn by atomic absorption spectroscopy (PerkinElmer 1100).

157 The DOP index (deviation from optimum percentage) was estimated for the diagnosis
158 of the nutritive status of the trees (Montañés et al., 1993). This index provides similar
159 information to the Diagnosis and Recommendation Integrated System (DRIS) (Sanz,
160 1999). The DOP index was calculated from the leaf analysis by the following
161 mathematical expression:

$$DOP = \frac{C \times 100}{C_{ref}} - 100$$

162 where C is the nutrient concentration in the sample to be studied and C_{ref} is the nutrient
163 concentration considered as optimum, both values given on a dry matter basis. The C_{ref}
164 has been taken from optimum values proposed by Leece (1975). The ΣDOP is obtained
165 by adding the values of DOP indices irrespective of sign. The larger was the ΣDOP the
166 greater was the intensity of imbalances among nutrients.
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170 2.6. Data analysis

171 Data were evaluated by two-way variance (ANOVA) analysis with the program SPSS
172 21.0 (SPSS, Inc, Chicago, USA). When the F test was significant, means were separated
173 by Duncan's multiple range test ($P \leq 0.05$). A principal component analysis (PCA) was
174 performed to understand how agronomic and leaf traits contribute to variability among
175 the different rootstocks budded with 'Big Top' nectarine cultivar, using Unscrambler X
176 10.3 software (CamoAsa, 2001).

177 **3. Results and discussion**

178 3.1. Tree mortality

179 At the thirteenth year after budding, replant and heavy soil conditions generated varying
180 levels of tree mortality (Fig. 1). Adafuel, Garnem and Monegro rootstocks experienced
181 the highest tree mortality with 80%, 60% and 80% of dead trees, respectively.
182 Therefore, they were excluded of the study. Poor adaptation of Garnem and Felinem to
183 heavy soil conditions was already mentioned by Zarrouk et al. (2005). However, better
184 adaptation of Adafuel budded with different peach and nectarine cultivars was reported
185 in other studies (Font i Forcada et al., 2012; Zarrouk et al., 2005). For Adarcias,
186 Adesoto, Barrier, Cadaman, GF 677 and Mayor, only one tree per rootstock was lost.
187 Some authors considered Adesoto (Massai and Loreti, 2004) and GF 677 tolerant
188 rootstocks to replant conditions. Indeed, Adesoto was released because it adapts well to
189 highly calcareous and heavy soils, being tolerant to root asphyxia and Fe chlorosis
190 (Moreno et al., 1995a). However, in this study both Adesoto and GF 677 experienced a
191 20% mortality rate, in agreement with results obtained by Jiménez et al. (2011). In
192 contrast, all trees budded on 'Replantpac' survived and seem to tolerate better replant
193 and heavy soil conditions.

194 In the growing conditions, tree mortality could be attributed to the sensitivity of some
195 almond \times peach hybrid rootstocks to root asphyxia caused by waterlogging (Felipe,
196 2009) or susceptibility to various root rot pathogens such as *Phytophthora* spp. (Zarrouk
197 et al., 2005). Soil conditions and flooding irrigation are prone to waterlogging. The
198 soilborne fungi *Rosellinia necatrix* Prill and *Armillaria mellea* Vahl. P. are associated
199 with a high mortality rate in replant sites where peach-almond hybrids are used as
200 rootstocks in Spain (Jiménez et al., 2011; Pinochet, 2010). However, the presence of
201 both pathogens has not been detected in the present work.

202

203 3.2. *Tree growth and yield characteristics*

204 The vegetative growth of trees, expressed as TCSA, showed a considerable influence
205 attributable to the rootstock as early as the fourth year of growth (Fig. 2). At the
206 thirteenth year of scion growth, 'Big Top' showed higher TCSA values on Cadaman
207 and Replantpac (356.7 cm² and 342.2 cm², respectively), compared with the plums
208 Adesoto and Tetra, and the peach-almond Adarcias (173.6 cm², 204.7 cm² and 207.1
209 cm², respectively). On Adarcias, Adesoto and Tetra, the reduction in TCSA was 42%,
210 52% and 43% compared to Cadaman, and 40%, 50% and 41% compared to Replantpac.
211 Tree growth was intermediate on the other rootstocks (Table 2 and Fig. 2). The low-
212 medium vigour of Adarcias and Tetra and the high vigour of Cadaman have already
213 been mentioned (Font i Forcada et al., 2012; Hudina et al., 2006; Moreno et al., 1994).
214 In a different replanting soil, Adesoto and Tetra resulted in a medium vigour (Jiménez
215 et al., 2011). Mayor, Barrier, GF 677 and Felinem showed an intermediate TCSA and
216 around 20% and 23% reduction in trunk size compared to Replantpac and Cadaman,
217 respectively. However, Zarrouk et al. (2005) and Font i Forcada et al. (2012) reported
218 Felinem as one of the most vigorous rootstocks in similar soil conditions, but not under
219 replant conditions.

220 In the first bearing years (2003 and 2004), yields were insignificant, and there were no
221 statistically significant differences among rootstocks. However, in the following
222 cropping years differences among rootstocks became evident (Mestre, 2012). In 2013,
223 Cadaman showed the greatest cumulative yield although it did not differ from Barrier
224 and Replantpac. The highest yield and cumulative yield efficiency of Cadaman were
225 already mentioned (Massai and Loreti; 2004; Zarrouk et al., 2005). The lowest
226 cumulative yield was recorded on the less vigorous rootstocks (Adarcias, Adesoto and
227 Tetra) but did not differ from Felinem, GF 677 and Mayor.

228 'Big Top' budded on GF 677 showed the highest yield efficiency, although differences
229 were not significant when compared with Adarcias, Adesoto, Barrier, Felinem and
230 Tetra. The lowest yield efficiency was recorded on Mayor, although not significantly
231 different from Adesoto, Cadaman, Felinem, Replantpac and Tetra (Table 2). Thus, less
232 vigorous rootstocks (Adarcias, Adesoto and Tetra) induced yield efficiency similar to
233 that on more invigorating rootstocks as Cadaman, GF 677 and Replantpac.

234 The average of fruit weight for the last five years of study was significantly affected by
235 rootstocks, as observed during the previous years (Mestre, 2012). The peach × *P.*
236 *davidiana* Barrier, with an intermediate level of vigour, tended to show higher fruit

237 weight, especially when compared with the peach-almond hybrids Felinem and Mayor
238 (Table 2). The tendency of Barrier to induce higher fruit weight has been previously
239 reported (Loreti and Massai, 2006; Orazem et al., 2011).

240 Yield was generally proportional to growth or tree size. Thus, positive correlations were
241 found between rootstock vigour and cumulative yield ($r=0.84$; $P\leq 0.05$) in 2013, and
242 between annual yield and vigour, except in the period 2003-2006 (Mestre, 2012).
243 However, the greater vigour on fertile and well-irrigated soils may become excessive
244 for good orchard practice unless some irrigation and other cultural practices are
245 modified (Font i Forcada et al., 2012). Vigorous rootstocks appear suitable for peach
246 production under harsh replant conditions or in poor and calcareous soils that might
247 otherwise be unfavorable for growing peach (Cambra, 1990; Moreno et al., 1994;
248 1996). Replantpac and Cadaman were the most vigorous rootstocks and seem to induce
249 a higher cumulative yield, demonstrating a good adaptation to the growing conditions.
250 In contrast, Adarcias, Adesoto and Tetra with lower vigour and medium to high yield
251 efficiency may be suitable for reducing excessive growth of peach cultivars or to
252 increase planting density (Moreno and Cambra, 1994) allowing the possibility of
253 establishing pedestrian orchards with the benefits of reducing labour costs, especially at
254 pruning and harvest (Jiménez et al., 2011).

255 *3.4. Root suckering*

256 The number of suckers per tree was also determined. The Pollizo Adesoto consistently
257 showed the highest number of root suckers, as previously described by Moreno et al.
258 (1995a) and Reighard et al. (2008). Excessive rootstock suckering is a common
259 drawback observed with some plums (Reighard et al., 1997, 2008; Salesses et al., 1998).
260 A fewer number of suckers, nearly always in the form of crown suckers, were observed
261 for Barrier, Cadaman, Felinem, GF 677 and Replantpac. Adarcias and Tetra did not
262 produce suckers during all the cropping years (Table 2), as reported by Nicotra and
263 Moser (1997) for Tetra.

264 *3.5. Leaf chlorophyll content*

265 Leaf SPAD readings were higher for Replantpac, although it did not differ from Barrier,
266 Cadaman, Felinem and Mayor (Table 3). Lower values were found on Adarcias,
267 although differences were not significant when compared with Adesoto, Barrier,
268 Cadaman, GF 677, Mayor and Tetra. However, Jiménez et al. (2008) classified Adesoto
269 and GF 677 as tolerant to iron chlorosis according to their capacity to reduce iron from

270 the soil. SPAD values were in the same range as previously reported (Jiménez et al.,
271 2011; Zarrouk et al., 2005).

272 *3.6. Leaf mineral nutrients and DOP index*

273 The results showed that most nutrients were affected by the choice of rootstock (Table
274 4). Leaf N concentration was higher on Cadaman but not different from GF 677 and
275 Replantpac, and lower on Adarcias and Felinem although differences were not
276 significant from Adesoto, Barrier, Mayor and Tetra. The tendency of Cadaman and GF
277 677 to show higher leaf N concentration than Adarcias and Felinem was also described
278 by Zarrouk et al. (2005) with different cultivars. All rootstocks showed N deficiency,
279 according to reference values (Leece et al., 1975), but comparable N concentration has
280 been previously reported (Zarrouk et al., 2005) in similar growing conditions. The P
281 concentration was higher on Adarcias, whereas it was lower on Adesoto and Cadaman,
282 although it did not differ when compared with the other rootstocks. Nevertheless, leaf P
283 concentrations were considered to be adequate for all rootstocks according to Leece
284 (1975). In similar soil conditions, Adarcias showed lower P values than Cadaman
285 (Zarrouk et al., 2005). The highest leaf K concentration was obtained on the plums
286 Adesoto and Tetra, showing values higher than optimum (Leece, 1975). They were
287 followed by the plum-almond Replantpac with optimum values. In contrast, Adarcias
288 and Felinem induced lower values, but they did not differ significantly from Cadaman
289 and Mayor, all of them presenting marginal values (Leece, 1975). It is interesting to
290 note that plum-based rootstocks increase K uptake more than peach-based rootstocks in
291 these soil conditions. Other authors reported K deficiency for peach (Zarrouk et al.,
292 2005) and cherry cultivars (Jiménez et al., 2007; Moreno et al., 1996, 2001) grown in
293 calcareous soils probably due to poor uptake of this element in this type of soils.
294 Johnson and Uriu (1989) reported that lower leaf K concentration is probably due to its
295 fixation by clay particles in the soil. The clay-loam texture in our growing conditions
296 could also explain the lower leaf concentration of K for some of the evaluated
297 rootstocks, especially for peach-based rootstocks. Leaf Ca concentration was higher on
298 Adarcias, Felinem, GF 677 and Tetra than on the other rootstocks, showing values
299 slightly higher than optimum (Leece, 1975). The higher leaf Ca concentration of trees
300 budded on Felinem has been previously reported (Zarrouk et al., 2005). The Mg
301 concentration was higher on Barrier and lower on Adesoto and Tetra, although it did not
302 differ from the other rootstocks, exhibiting all of them optimum values (Leece, 1975)
303 with the exception of Tetra. The low values of Mg in the case of Tetra could be

304 explained by the antagonism of Mg with some elements such as Ca in heavy-calcareous
305 soil conditions (Zarrouk et al., 2005).

306 Lower leaf macronutrients concentration has been reported in less vigorous rootstocks
307 for peach (Zarrouk et al., 2005), cherry (Moreno et al., 2001), and apricot (Rosati et al.,
308 1997), suggesting that dwarfing rootstocks could be less efficient in the absorption of
309 some macronutrients from the soil.

310 The highest Fe concentration was shown on Adesoto, and the lowest on the peach \times *P.*
311 *davidiana* Barrier and Cadaman, but they did not differ significantly from the other
312 rootstocks. The performance of Adesoto to induce higher Fe concentration than other
313 rootstocks shows the interest of Adesoto in calcareous soils. Nevertheless, all rootstocks
314 presented lower Fe concentrations than the optimum according to Leece (1975),
315 especially Barrier as previously reported by Jiménez et al. (2008). Low iron
316 bioavailability is mainly the result of its insolubility at higher pH values, especially in
317 calcareous soils, where roots of some species are unable to acquire Fe (Hell and
318 Stephan, 2003). Most tolerant rootstocks to iron chlorosis are, in general, *P. amygdalus*
319 \times *P. persica* hybrids. However, plum rootstocks (Adesoto and Tetra) did also appear to
320 be more tolerant to iron-chlorosis than *P. persica* \times *P. davidiana* rootstocks. The Cu
321 concentration was higher on Adesoto, and lower on Barrier, Cadaman and Tetra,
322 although they did not differ from the other rootstocks. Leaf Cu concentration was
323 adequate for all rootstocks, except for Adesoto with slightly higher values compared to
324 the optimum (Leece, 1975). The highest Mn concentration was observed on Replantpac,
325 although it did not differ from Mayor. According to reference values (Leece, 1975), all
326 rootstocks showed Mn values lower than optimum except Replantpac. Mn deficiency
327 has been also reported for peach and cherry grown in calcareous soils (Jiménez et al.,
328 2004; Moreno et al., 1996, 2001; Zarrouk et al., 2005) probably due to the
329 insolubilization of this element in this type of soil. Furthermore, increased Ca in soil or
330 an excess of phosphoric acid fertilization might decrease or block Mn uptake (Johnson
331 and Uriu, 1989). Zarrouk et al. (2005) reported significant correlations between
332 chlorophyll concentration and K and Mn leaf concentration. The performance of
333 Replantpac to show higher SPAD, and K and Mn values is related with the role that Mn
334 plays in the photosynthesis process, and K in the tree vegetative development as was
335 reported for cherry (Jiménez et al., 2004). The highest Zn concentration was found on
336 Tetra, but not significantly different from Adesoto and Barrier, showing Adesoto and
337 Tetra optimum values (Leece, 1975).

338 According to the Σ DOP index, Barrier and Cadaman showed wider imbalanced
339 nutritional values, whereas Tetra showed the best balanced in nutritional values,
340 although it did not differ from GF 677 and Mayor.

341 3.7. Principal component analysis

342 The first two PCs (PC1 and PC2) accounted 59% of the total variance (Fig. 3). PC1
343 represented 36% of the variance and PC2 showed the 23% of the variance. Main
344 sources of variability with the highest Eigen vectors in each PC were as follows. PC1:
345 DOP, Fe, Ca, TCSA, CY, and Mg; PC2: suckering, K, Cu and P. The results of the
346 analysis of PCA showed that rootstocks on the negative side of PC1 corresponding to
347 Replantpac, Cadaman and Barrier induced in general higher TCSA, CY and leaf Mg
348 concentration, and lower DOP and Fe and Ca leaf concentrations, whereas Adesoto and
349 Tetra induced the contrary. In addition, Replantpac showed higher values on FW, N and
350 SPAD. Rootstocks on the positive side of PC1 and PC2, including Adesoto, showed
351 higher suckering values and higher K and Cu leaf concentrations, and lower values of
352 DOP, P and Ca.

353 The results obtained with the PCA confirm that the good adaptation of Replantpac to
354 the growing conditions probably favoured higher vigour, cumulative yield and fruit
355 weight, as well as higher N leaf content and SPAD values.

356 4. Conclusions

357 Performance of 'Big Top' nectarine was influenced by *Prunus* rootstock adaptive
358 capacity to the growing conditions. The choice of rootstock requires careful analysis of
359 the interaction between graft combination and agronomic characteristics of the area
360 considered.

361 In replant and heavy-calcareous soil conditions, the best adaptation of Replantpac is
362 highlighted by the absence of dead trees, thirteenth years after planting, especially when
363 compared with the peach-almond hybrids Adafuel, Garnem and Monegro, likely more
364 susceptible to root asphyxia. In addition, Replantpac rootstock appears suitable for
365 peach production when planting on marginal soils or under replanting conditions,
366 showing higher vigour and cumulative yields. In contrast, the lower vigour and good
367 yield efficiency of Adarcias, Adesoto and Tetra rootstocks may be suitable for reducing
368 excessive growth of peach cultivars in fertile soils and to increase planting density.

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375 **6.References**

376 Bellini, E., Nencetti, V., Natarelli, L., Liverani, A., Insero, O., Conte, L., 2004. Liste
377 varietali dei fruttiferi 2004: Pesco. L'Informatore Agrario 24, 53–69.

378 Cambra, R., 1990. Adafuel, an almond x peach hybrid rootstock. HortScience 25, 584–
379 584.

380 CamoASA, 2001. The unscrambler 9.6 user manual. CamoASA, Oslo.

381 C.I.I. (Comité Inter-Institutos para el estudio de técnicas analíticas), 1969. Métodos de
382 referencia para la determinación de elementos minerales en vegetales. Anales de
383 Edafología y Agrobiología 38, 403–417.

384 C.I.I. (Comité Inter-Institutes), Pinta, M., DeWaele, G. 1975. Etalons végétaux pour
385 l'analyse foliaire. In: Kozma, P., (Ed.), Le Contrôle de l'Alimentation des Plantes
386 Cultivées. Akadémiai Kiadó, Budapest, pp. 159–172.

387 Cos, J., Frutos, D., García, R., Rodríguez, J., Carrillo, A., 2004. In Vitro rooting study
388 of peach almond hybrid Mayor[®]. Acta Hort. 658, 623–628.

389 Della Strada, G., Fideghelli, C., 2003. L'aggiornamento varietale in peschicoltura. Riv.
390 Frutticolt. 7–8, 8–12.

391 FAOSTAT, 2014. (<http://faostat.fao.org>)

392 Felipe, A.J., 2009. Felinem, Garnem, and Monegro almond x peach hybrid rootstocks.
393 HortScience 44, 196–197.

394 Fernández, C., Pinochet, J., Esmenjaud, D., Salesses, G., Felipe, A., 1994. Resistance
395 among new *Prunus* rootstocks and selections to root-knot nematodes in Spain and France.
396 HortScience 29, 1064–1067.

397 Font i Forcada, C., Gogorcena, Y., Moreno, M.A., 2012. Agronomical and fruit quality
398 traits of two peach cultivars on peach-almond hybrid rootstocks growing on
399 Mediterranean conditions. Sci. Hortic. 140, 157–163.

400 Font i Forcada C., Gogorcena, Y., Moreno, M.A., 2014. Agronomical parameters, sugar
401 profile and antioxidant compounds of “Catherine” peach cultivar influenced by different
402 plum rootstocks. Int. J. Mol. Sci. 15(2), 2237-2254.

403 Giorgi, M., Capocasa, F., Scalzo, J., Murri, G., Battino, M., Mezzetti, B., 2005. The
404 rootstock effects on plant adaptability, production, fruit quality, and nutrition in the
405 peach (cv 'Suncrest'). Sci. Hortic. 107, 36–42.

406 Hell, R., Stephan, U.W., 2003. Iron uptake, trafficking and homeostasis in plants. *Planta*
407 216, 541–551.

408 Hudina, M., Fajt, N., Štampar, F., 2006. Influence of rootstock on orchard productivity
409 and fruit quality in peach cv. ‘Redhaven’. *J. Hortic. Sci. Biotechnol.* 81, 1064–1068.

410 Iglesias, I., 2010. La coltivazione del pesco in Spagna: situazione produttiva,
411 innovazioni varietali e tecniche colturali. *Italus Hortus* 17 (3), 7–10.

412 Jiménez, S., Garín, A., Gogorcena, Y., Betrán, J.A., Moreno, M.A., 2004. Flower and
413 foliar analysis for prognosis of sweet cherry nutrition: influence of different rootstocks.
414 *J. Plant. Nutr.* 27, 701–712.

415 Jiménez, S., Pinochet, J., Gogorcena, Y., Betrán, J.A., Moreno, M.A., 2007. Influence
416 of different vigour cherry rootstocks on leaves and shoots mineral composition. *Sci.*
417 *Hortic.* 112, 73–79.

418 Jiménez, S., Pinochet, J., Abadía, A., Moreno, M.A., Gogorcena, Y., 2008. Tolerance
419 response to iron chlorosis of *Prunus* selections as rootstocks. *HortScience* 43, 304–309.

420 Jiménez, S., Pinochet, J., Romero, J., Gogorcena, Y., Moreno, M.A., Espada, J.L., 2011.
421 Performance of peach and plum based rootstocks of different vigour on a late peach
422 cultivar in replant and calcareous conditions. *Sci. Hortic.* 129, 58–63.

423 Johnson, R.S., Uriu, K., 1989. Mineral nutrition. In: Larue, J., Johnson, R.S. (Eds.),
424 *Growing and Handling for Fresh Market: Division of Agriculture Resource.* University
425 of California, Oakland, CA, pp. 68–81.

426 Leece, D.R., 1975. Diagnostic leaf analysis for stone fruit. Peach (*Prunus persica*).
427 *Aust. J. Exp. Agric. Anim. Husb.* 15, 138–139.

428 Loreti, F., Massai, R., 2006. Bioagronomic evaluation of peach rootstocks by the Italian
429 MiPAF targeted Project. *Acta Hort.* 713, 295–302.

430 MAGRAMA, 2014. <<http://www.magrama.es>>.

431 Massai, R., Loreti, F., 2004. Preliminary observations on nine peach rootstocks grown
432 in a replant soil. *Acta Hort.* 658, 185–192.

433 Mestre, L., 2012. Agronomic performance of peach rootstocks. Influence on mineral
434 nutrition and fruit quality. Final degree Project and Dissertation, 76 pp. December 13th,
435 2012. B.S. in Agricultural Engineering. EUPLA, University of Zaragoza, Spain.

436 Montañés, L., Heras, L., Abadía, J., Sanz, M., 1993. Plant analysis interpretation based
437 on a new index: deviation from optimum percentage (DOP). *J. Plant Nutr.* 16, 1289–
438 1308.

439 Moreno, M.A., Cambra, R., 1994. Adarcias - an almond × peach hybrid rootstock.
440 HortScience 29, 925–925.

441 Moreno, M.A., Tabuenca, M.C., Cambra, R., 1994. Performance of Adafuel and
442 Adarcias as peach rootstocks. HortScience 29, 1271–1273.

443 Moreno, M.A., Tabuenca, M.C., Cambra, R., 1995a. Adesoto 101, a plum rootstock for
444 peaches and other stone fruits. HortScience 30, 1314–1315.

445 Moreno, M.A., Tabuenca, M.C., Cambra, R., 1995b. Adara, a plum rootstock for
446 cherries and other stone fruit species. HortScience 30, 1316–1317.

447 Moreno, M.A., Montañés, L., Tabuenca, M.C., Cambra, R., 1996. The performance of
448 Adara as a cherry rootstock. Sci. Hortic. 65, 85–91.

449 Moreno, M.A., Adrada, R., Aparicio, J., Betrán, J.A., 2001. Performance of ‘Sunburst’
450 sweet cherry grafted on different rootstocks. J. Hortic. Sci. Biotechnol. 76, 167–173.

451 Nicotra, A., Moser, L., 1997. Two new plum rootstocks for peach and nectarines: Penta
452 and Tetra. Acta Hort. 451, 269–271.

453 Orazem, P., Stampar, F., Hudina, M., 2011. Quality analysis of ‘Redhaven’ peach fruit
454 grafted on 11 rootstocks of different genetic origin in a replant soil. Food Chem. 124,
455 1691–1698.

456 Pinochet, J. 1997. Breeding and selection for resistance to root-knot and lesion nematodes
457 in *Prunus* rootstocks adapted to Mediterranean conditions. Phytoparasitica 25, 271–274.

458 Pinochet, J., 2009. ‘Greenpac’, a new peach hybrid rootstock adapted to Mediterranean
459 conditions. HortScience 44, 1456–1457.

460 Pinochet, J., 2010. ‘Replantpac’ (Rootpac®), a plum-almond hybrid rootstock for
461 replant situations. HortScience 45, 299–301.

462 Reighard, G.L., Newall W.C., Beckman T.G., Okie W.R., Zehr E.I., Nyczepir A.P.,
463 1997. Field performance of *Prunus* rootstock cultivars and selections on replant soils in
464 South Carolina. Acta Hort. 451, 243–249.

465 Reighard, G.L., Ouellette, D., Brock, K., 2008. Performance of new *Prunus* rootstocks
466 for peach on South Carolina. Acta Hort. 772, 237–240.

467 Remorini, D., Tavarini, S., Degl’Innocenti, E., Loreti, F., Massai, R., Guidi, L., 2008.
468 Effect of rootstocks and harvesting time on the nutritional quality of peel and flesh of
469 peach fruits. Food Chem. 110, 361–367.

470 Rosati, A., DeJong, T.M., Southwick, S.M., 1997. Comparison of leaf mineral content,
471 carbon assimilation and stem water potential of two apricot (*Prunus armeniaca*)
472 cultivars grafted on ‘Citation’ and ‘Mariana 2624’ rootstocks. Acta Hort. 451, 263–267.

473 Sanz, M., 1999. Evaluation of interpretation of DRIS system during growing season of
474 the peach tree: comparison with DOP method. *Commun. Soil Sci. Plant Anal.* 30 (7 and
475 8), 1025–1036.

476 Saleses, G., Dirlewanger, E., Bonnet, A., Lecouls, A.C., Esmenjaud, D., 1998.
477 Interspecific hybridization and rootstock breeding for peach. *Acta Hort.* 465, 209–217.

478 Zarrouk, O., Gogorcena, Y., Gómez-Aparisi, J., Betrán, J.A., Moreno, M.A., 2005.
479 Influence of almond x peach hybrids rootstocks on flower and leaf mineral
480 concentration, yield and vigour of two peach cultivars. *Sci. Hortic.* 106, 502–514.

Table 1

List of studied rootstocks, description and origin

Rootstock	Species	Genetic background	Origin ^a	References
Adafuel	<i>P. amygdalus</i> × <i>P. persica</i>	‘Marcona’ seedlings (open-pollinated)	CSIC, Spain	Cambra (1990)
Adarcias	<i>P. amygdalus</i> × <i>P. persica</i>	Open-pollinated	CSIC, Spain	Moreno and Cambra (1994); Moreno et al. (1994)
Adesoto	<i>P. insititia</i>	Open-pollinated	CSIC, Spain	Moreno et al. (1995a)
Barrier	<i>P. davidiana</i> × <i>P. persica</i>	Open-pollinated	ISF, Italy	De Salvador et al. (2002)
Cadaman	<i>P. davidiana</i> × <i>P. persica</i>	Controlled cross	INRA (France- Hungary)	Edin and Garcin (1994)
Felinem	<i>P. amygdalus</i> × <i>P. persica</i>	‘Garfi’ almond × ‘Nemared’ peach	CITA, Spain	Felipe (2009)
Garnem	<i>P. amygdalus</i> × <i>P. persica</i>	‘Garfi’ almond × ‘Nemared’ peach	CITA, Spain	Felipe (2009)
Monegro	<i>P. amygdalus</i> × <i>P. persica</i>	‘Garfi’ almond × ‘Nemared’ peach	CITA, Spain	Felipe (2009)
GF 677	<i>P. amygdalus</i> × <i>P. persica</i>	Open-pollinated	INRA, France	Bernhard and Grasselly (1981)
Mayor	<i>P. amygdalus</i> × <i>P. persica</i>	Open-pollinated	CIDA, Spain	Cos et al. (2004)
Replantpac	<i>P. cerasifera</i> × <i>P. amygdalus</i>	Open-pollinated	AI, Spain	Pinochet (2010)
Tetra	<i>P. domestica</i>	Open-pollinated	ISF, Italy	Nicotra and Moser (1997)

^a AI = Agromillora Iberia S.L. private nursery, Spain; CITA = Centro de Investigación y Tecnología Agroalimentaria de Aragón; CIDA = Centro de Investigación y Desarrollo Agroalimentario de Murcia; ISF = Istituto Sperimentale per la Frutticoltura di Roma; CSIC = Consejo Superior de Investigaciones Científicas; INRA = Institut National de la Recherche Agronomique.

Table 2

Trunk cross-sectional area (TCSA), cumulative yield, yield efficiency and root or crown suckering of 'Big Top' budded on different rootstocks, at the thirteenth year after planting (2013). Mean values (2009-2013) of fruit weight.

Rootstock	TCSA (cm ²)	CY (kg tree ⁻¹)	YE (kg cm ⁻²)	SCK (suckers tree ⁻¹)	AFW (g)
Adarcias	207.1 ab	156.8 abc	0.76 bc	0.0 a	202.6 ab
Adesoto	173.6 a	130.3 a	0.75 abc	7.2 b	212.6 ab
Barrier	272.2 abc	218.9 bc	0.80 bc	1.0 a	226.7 b
Cadaman	356.7 c	230.8 c	0.65 ab	2.0 a	217.4 ab
Felinem	259.3 ab	155.1 abc	0.70 abc	0.3 a	188.2 a
GF 677	263.7 abc	207.4 abc	0.84 c	0.2 a	210.6 ab
Mayor	292.4 bc	170.5 abc	0.56 a	0.8 a	192.6 a
Replantpac	342.2 c	217.9 bc	0.63 ab	1.2 a	214.7 ab
Tetra	204.7 ab	138.2 ab	0.66 abc	0.0 a	210.9 ab

For each rootstock means followed by the same letter in each column are not significantly different at $P < 0.05$ according to Duncan's Multiple Range Test.

Abbreviations: AFW, average fruit weight; CY, cumulative yield; SCK, root and crown suckering; TCSA, trunk cross sectional area; YE, Yield efficiency.

1 **Table 3**

2 Effect of rootstock on leaf chlorophyll concentration, measured as SPAD values, of
3 'Big Top' nectarine cultivar, at the twelfth year after planting (2012).

Rootstock	SPAD
Adarcias	37.3 a
Adesoto	38.0 ab
Barrier	38.2 abc
Cadaman	38.2 abc
Felinem	39.3 bc
GF 677	37.7 ab
Mayor	38.6 abc
Replantpac	39.7 c
Tetra	37.7 ab

4 Means followed by the same letter in each column are not
5 significantly different at $P \leq 0.05$ according to Duncan's
6 multiple rang test.

7

Table 4

Rootstock effects on leaf mineral element concentrations of 'Big Top' at 120 days (110 D) after full bloom, by the twelfth year after planting (2012). Results for N, P, K, Ca and Mg are expressed as percentage of dry matter and for Fe, Mn, Cu and Zn, as mg kg⁻¹.

Rootstock	N	P	K	Ca	Mg	Fe	Cu	Mn	Zn	∑ DOP
Adarcias	2.4 a	0.21 b	1.6 a	2.8 e	0.37 abc	76.9 abc	7.4 ab	32.6 a	12.1 ab	- 276.2 bcd
Adesoto	2.5 abc	0.15 a	3.3 d	1.9 cd	0.30 ab	86.3 c	19.5 b	30.6 a	20.0 ef	- 271.7 bcd
Barrier	2.4 ab	0.16 ab	1.9 b	1.1 ab	0.47 c	59.8 a	8.2 a	26.2 a	19.4 ef	- 337.6 d
Cadaman	2.8 d	0.15 a	1.9 ab	1.6 bc	0.37 abc	67.3 ab	8.0 a	33.7 a	16.5 cd	- 320.8 d
Felinem	2.4 a	0.17 ab	1.3 a	2.9 e	0.38 abc	83.4 bc	8.5 ab	29.3 a	14.3 bc	- 298.4 bc
GF 677	2.7 bcd	0.16 ab	1.8 b	3.0 e	0.37 abc	73.1 abc	9.3 ab	31.7 a	18.2 de	- 217.9 ab
Mayor	2.5 abc	0.20 ab	1.5 ab	2.4 de	0.36 abc	83.5 bc	12.4 ab	37.0 ab	17.9 de	- 242.2 abc
Replantpac	2.7 cd	0.17 ab	2.8 c	0.6 a	0.39 abc	76.9 abc	10.2 ab	46.1 b	11.1 a	- 290.6 cd
Tetra	2.6 abc	0.18 ab	3.5 d	3.0 e	0.24 a	83.2 bc	7.3 a	35.5 a	21.6 f	- 191.3 a

Means followed by the same letter in each column are not significantly different at $P \leq 0.05$ according to Duncan's multiple range test.

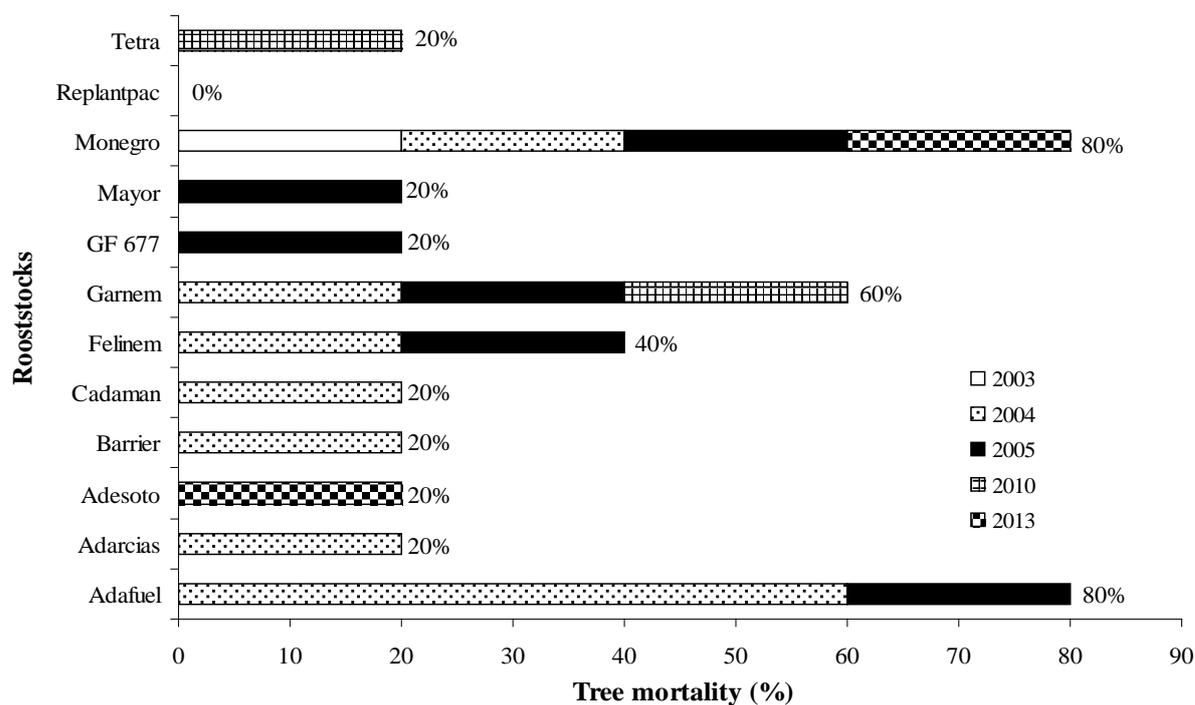


Fig. 1. Tree mortality rate (%) from the third (2003) to the thirteenth (2013) year after planting in the orchard trial. Percentages values right side of the bars indicated accumulated mortality rate at the end of the experiment.

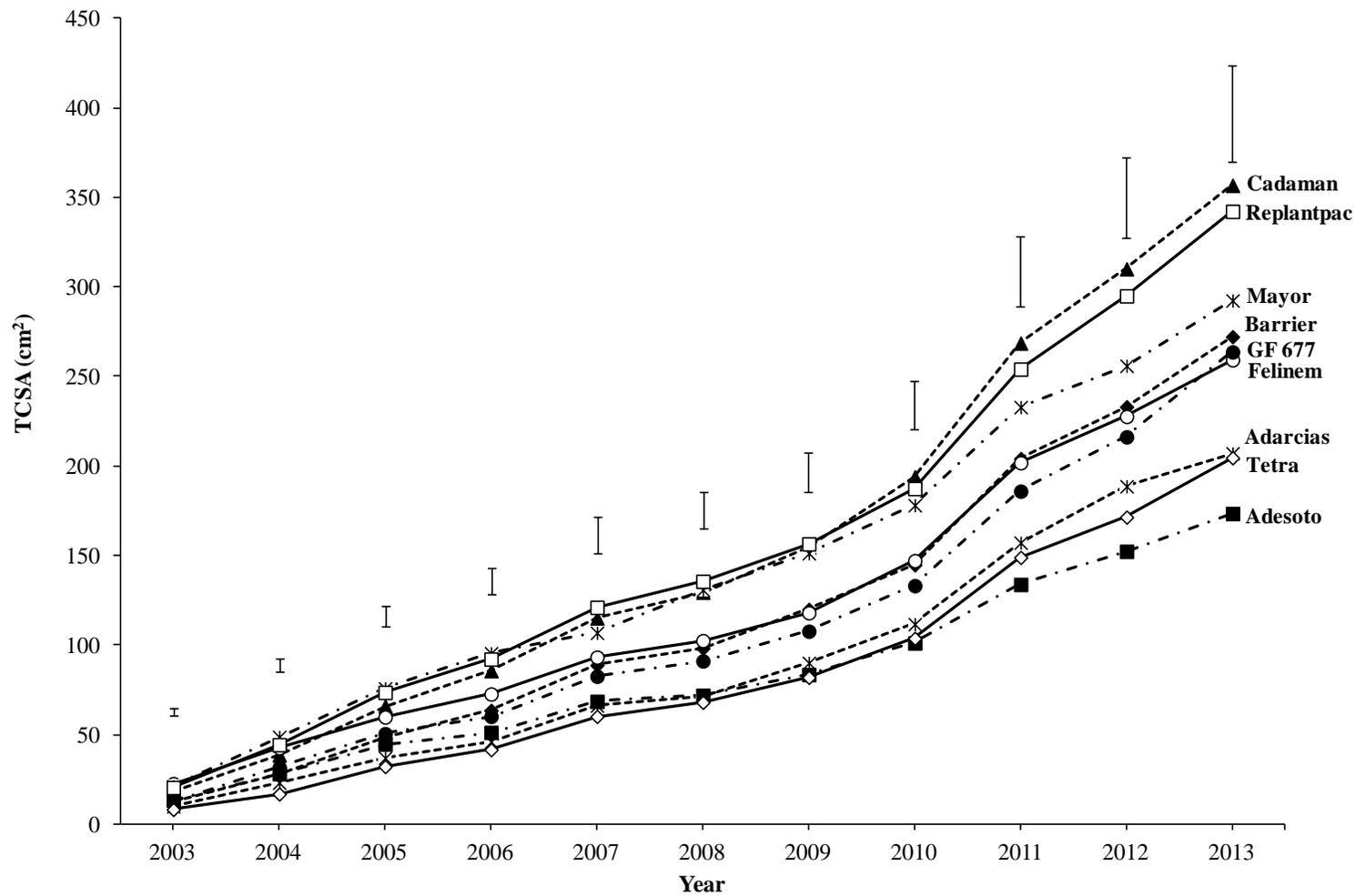


Fig. 2. Rootstock effects on TCSA (cm²) of 'Big Top' cultivar from the third (2003) to the thirteenth (2013) year after planting in the orchard. Vertical lines indicate LSD (P≤0.05)

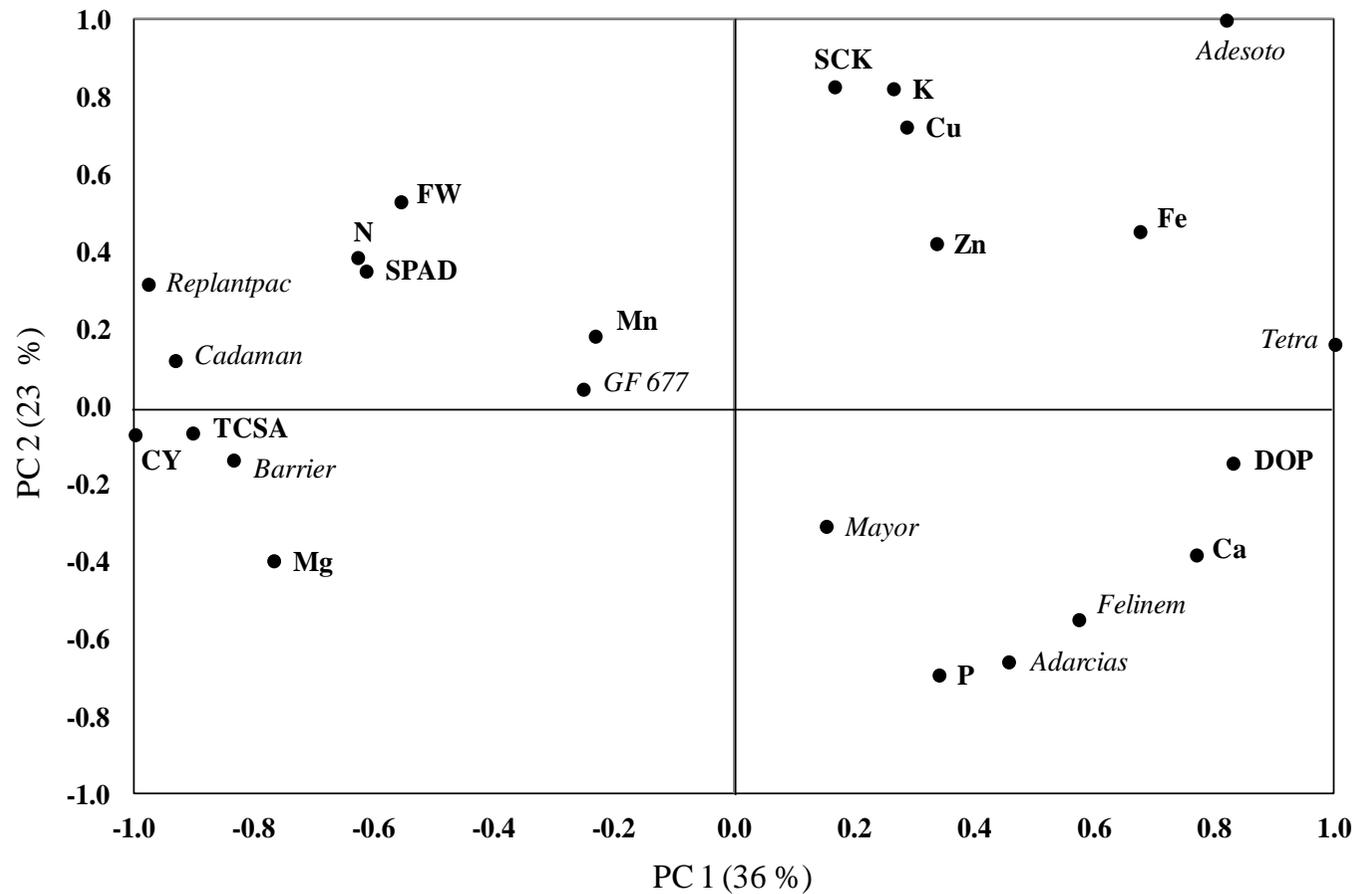


Fig. 3. Principal component analysis for agronomic and leaf traits evaluated on different rootstocks budded with ‘Big Top’ nectarine cultivar. Abbreviations: Cu, copper; CY, cumulative yield; DOP, deviation from optimum percentage index; Fe, iron; FW, fruit weight; K, potassium; Na, sodium; Mg, magnesium; Mn, manganese; N, nitrogen; P, phosphorus; SCK, root and crown suckering, TCSA, trunk cross sectional area; Zn, zinc.