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1 **Genotypic variability in radial resistance to water flow in olive roots and its response to**
2 **temperature variations**

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12 **Abstract**

13 As radial root resistance (R_p) represents one of the key components of the soil-plant-atmosphere
14 continuum (SPAC) resistance catena modulating water transport, understanding its control is
15 essential for physiologists, modelers and breeders. Reports of R_p , however, are still scarce and
16 scattered in the scientific literature. In this study we assessed genetic variability in R_p and its
17 dependence on temperature in five widely-used olive cultivars. In a first experiment, cultivar
18 differences in R_p at 25 °C were evaluated from flow-pressure measurements in excised roots
19 and subsequent analysis of root traits. In a second experiment, similar determinations were
20 performed continually over 5 h periods in which temperature was gradually increased from 12
21 to 32 °C, enabling the assessment of R_p response to changing temperature. Despite some
22 variability, our results did not show statistical differences in R_p among cultivars in the first
23 experiment. In the second, cultivar differences in R_p were not significant at 12 °C, but they
24 became so as temperature increased. Furthermore, the changes in R_p between 12 and 32 °C
25 were higher than those expected by the temperature-driven decrease in water viscosity, with
26 the degree of that change differing among cultivars. Also, R_p at 25 °C reached momentarily in
27 the second experiment was consistently higher than in the first at that same, but fixed,
28 temperature. Overall, our results suggest that there is limited variability in R_p among the studied
29 cultivars when plants have been exposed to a given temperature for sufficient time.
30 Temperature-induced variation in R_p might thus be partly explained by changes in membrane
31 permeability that occur slowly, which explains why our values at 25 °C differed between
32 experiments. The observed cultivar differences in R_p with warming also indicate faster
33 acclimation of R_p to temperature changes in some cultivars than others.

34 **Introduction**

35 Since the pioneering work by van den Honert (1948), the movement of water through the soil-
36 plant-atmosphere-continuum (SPAC) is often treated as a catenary process that can be
37 modelled following an Ohm's law analogy (Tyree and Ewers 1991). Accordingly, the water
38 flux from soil to leaves is proportional to water potential gradient and inversely proportional
39 to hydraulic resistance to flow between the extremes of the pathway. Studying the factors that
40 determine the SPAC hydraulic resistance is therefore crucial for improving our understanding
41 of plant water relations.

42 The hydraulic resistance to water transport from the soil to the leaves can be decomposed into
43 four elements in series, namely, the soil (R_{soil} , from the soil to the root surface), radial (R_{p} , from
44 the root surface to root vascular bundles), vascular (R_{xy} , from the root xylem to the leaves) and
45 mesophyll (R_{mes} , from the leaf veins to the evaporation sites) resistances. The first component
46 of the catena, R_{soil} , is known to be strongly modulated by the soil matrix composition, by soil
47 water content and by the amount of roots; and is very low as compared to the other components
48 when the soil is wet (Campbell 1985). R_{xy} is the main component of the resistance catena on a
49 distance basis, but the high specialization of xylem conduits limits its magnitude as resistor
50 (Sperry 2003, Venturas et al. 2017). R_{xy} is mainly constrained by a number of anatomical and
51 physiological factors, although harsh environmental conditions (e.g. severe water stress) can
52 also result in the cavitation of xylem conduits. The extravascular plant resistances (R_{p} and R_{mes})
53 constitute the least-known components of the resistance catena despite probably being the most
54 important bottlenecks in the whole-plant hydraulic continuum (Tyree and Zimmerman 2002).
55 In this regard, R_{mes} has been suggested to account for up to 30 % of the whole-plant hydraulic
56 resistance in well-watered plants (Sack et al. 2003, Sack and Holbrook 2006, Scoffoni and Sack
57 2017) while experimental evidence has revealed values of R_{p} 2–20 times higher than those of
58 R_{xy} (Nobel and Sanderson 1984).

59 R_p represents the degree of impermeability of the different tissues arranged in series that form
60 the root cylinder (Steudle and Peterson 1998), and is determined by anatomical root traits.
61 However, R_p can vary over time due to modifications in membrane fluidity, aquaporin
62 activation and suberization, which can be triggered under certain environmental conditions
63 (Running and Reid 1980, Passioura 1988, Lee et al. 2005a). In this regard, it is well documented
64 that both soil water deficits (Rodriguez-Dominguez and Brodribb 2019) and low temperatures
65 can lead to large increases in R_p (e.g. Ameglio et al. 1994, Wan et al. 2001, Lee et al. 2005a).
66 In fact, some studies have highlighted that the inclusion of algorithms describing the
67 temperature effects on R_p can improve substantially the predictive power of SPAC models
68 (Mellander et al. 2006, García-Tejera et al. 2016). Besides, diurnal variations in R_p associated
69 to plant circadian rhythms have been described for some species (Henzler et al. 1999, Caldeira
70 et al. 2014).

71 So far, not much attention has been given to the characterization of intra-specific variability in
72 R_p . This topic is highly appealing as, in theory, two ideotypes differing in R_p might have
73 different patterns of water use along a season. A high R_p could imply an impaired water uptake
74 and slow plant growth, but it may still be a desirable trait in dry environments if the so-reduced
75 water uptake results in higher soil water availability during critical periods at the end of the
76 crop cycle. On the other hand, genotypic differences in R_p have been associated with differences
77 in chilling sensitivity (Aroca et al. 2001) and frost resistance (Pérez-López et al. 2010).

78 This paper deals with the evaluation of cultivar variability in R_p for olive trees. This evergreen
79 species is one of the most important crops in the Mediterranean basin, where they cover around
80 10.5 Mha (FAOSTAT 2017). Earlier studies on olive trees demonstrated that low temperatures
81 produce a disturbance of water relations, often evidenced by low water potentials occurring
82 even under low evaporative demand and adequate soil water content (Pavel and Fereres 1998,
83 Pérez-López et al. 2010, López-Bernal et al. 2015). This chilling-induced dehydration has been

84 reportedly linked to increases in R_p in olive and other sensitive species (Aroca et al. 2012,
85 Centeno et al. 2018). Indeed, García-Tejera et al. (2016) characterized the response of R_p to
86 temperature for the cultivar ‘Picual’, showing that the values measured at 10 °C were c.a. three
87 times higher than those measured at 25 °C.

88 The specific goals of the present study are the identification of cultivar differences 1) in R_p at
89 a mild and low temperatures and 2) in the response of R_p to short-term temperature variations.
90 In order to do so, two experiments were performed with rooted cuttings of five widely used
91 olive cultivars. In the first, we assessed cultivar variability in R_p at 25 °C, while the second
92 explored the differences at 12 °C and during a subsequent gradual increase of temperature up
93 to 32 °C.

94

95 **Materials and Methods**

96 *Plant material*

97 The experiments were conducted between June and July 2018 with rooted cuttings of five olive
98 cultivars, namely ‘Picual’, ‘Arbequina’, ‘Hojiblanca’, ‘Arbosana’ and ‘Frantoio’. Plant
99 material was obtained from a commercial nursery by early spring and subsequently grown
100 outdoor at the Institute for Sustainable Agriculture (IAS-CSIC, Córdoba, Spain) in small pots
101 filled with peat moss and irrigated meeting evapotranspiration every day.

102 *Experiment I*

103 Experiment I was aimed to assess cultivar differences in R_p at 25 °C. All the measurements
104 were performed inside a growth chamber with controlled temperature and a fixed 14-h
105 photoperiod with fluorescent lights at $360 \mu\text{mol m}^{-2} \text{s}^{-1}$. Both the plant material and instruments
106 were placed in the chamber 24 h prior to the start of measurements to ensure that everything

107 was at 25 °C by the beginning of the experiment. Five plants per cultivar were used for the
108 measurements.

109 The protocol followed for determining R_p was similar to that used by García-Tejera et al.
110 (2016). First, plants were extracted from their pots and immersed in water in order to remove
111 most part of the substrate, avoiding any injuries to the root systems. Then, the upper part of the
112 stem was cut 4 cm above the collar and the whole detached root system was placed in a pot
113 filled with water inside the vessel of a pressure chamber (Soil Moisture Equipment Corp., Santa
114 Barbara, CA, USA). The chamber was used to apply a constant 0.4 MPa pressure to root
115 systems for at least 45 min, in order to ensure a steady flux of xylem exudate (García-Tejera et
116 al. 2016). During this period, the xylem exudate was collected at 15 min intervals using cotton-
117 filled sample tubes. The average flux was determined by weighting the cotton-filled sample
118 tubes before and after each measurement period using a 0.0001 g precision balance (model
119 AV104, Mettler Toledo, Greifensee, Switzerland). Once flux determinations were completed,
120 root systems were washed with water to remove all the remaining substrate particles and then
121 scanned with a commercial scanner (HP Scanjet G3110). The WinRhizo software (Regent
122 Instruments Inc., Quebec City, QC, Canada) was used for estimating the values of several root
123 traits including average root diameter, root diameter frequency distribution and total root
124 surface (A). In all cases we assumed a maximum root diameter of 1.4 mm for absorbing roots
125 (Polverigiani et al. 2011, García-Tejera et al. 2016). Finally, an apparent value for the root
126 resistance (R_{root} , MPa m² s kg⁻¹) was estimated as:

$$127 \quad R_{root} = \Delta P A / F \quad (1)$$

128 Where ΔP is the pressure applied to the root system (0.4 MPa), A is root surface expressed in
129 m² and F , the flux of xylem exudate during the interval (kg s⁻¹). Because the hydraulic
130 resistance in the root xylematic pathway is generally considered negligible when compared to

131 the radial component (Nobel and Sanderson 1984, García-Tejera et al. 2016), and since our
132 rooted cuttings had small root systems, we assumed that $R_p = R_{root}$.

133 ***Experiment II***

134 The goal of Experiment II was to explore the responses of R_p to short-term changes in
135 temperature for the studied olive cultivars. In order to do so, we performed continuous
136 pressure-flux measurements similar to those of Experiment I over periods of five hours, during
137 which the temperature of the root systems increased from c.a. 12 to 32–35 °C. Three plants per
138 cultivar were used.

139 In the evening preceding the measurements (at 17.00 GMT), the plants were transferred to a
140 growth chamber with controlled temperature at 12 °C. Starting at around 7.00 GMT, flux
141 measurements were initially performed every 15 min inside the growth chamber with an
142 operation pressure of 0.4 MPa until the flux was steady. At such low temperature, we observed
143 that the time for the flux to stabilize was longer than that at 25 °C, taking between 60 and 120
144 min (Table S1). In all cases, only the data collected at 120 min were used in further analysis.
145 Then, the equipment was moved to an outdoor site exposed to direct sunlight. The 15-min flux
146 determinations continued there until around 12.00 GMT, with the rising solar radiation and air
147 temperature driving a gradual increase in the temperature of the root system, which was also
148 monitored every second with a thermocouple (Type E) placed inside the water-filled vessel
149 within the pressure chamber and controlled with a data logger (CR1000, Campbell Scientific
150 Inc., Logan, UT, USA). Following the pressure-flux measurements, root structural properties
151 were analyzed as in the previous experiment and values of R_p were calculated. Figure 1 shows
152 the typical patterns of temperature and estimated R_p found during the pressure-flow
153 measurements in Experiment II.

154 Statistical differences in R_p between cultivars were tested at various specific temperatures: 12,
155 16, 20, 24, 25, 28 and 32 °C. To do so, third degree polynomial functions were fitted to the
156 plots of R_p versus temperature for each replicate. Those fits were subsequently used for
157 estimating the values corresponding to the aforementioned temperatures.

158 In a further analysis, we assessed cultivar differences in the pattern and extent of the relative
159 decrease in R_p with temperature from the initial value measured at 12 °C. Such relative
160 resistance value was calculated for each replicate by dividing the estimated R_p by the value
161 inferred from the polynomial fit at 12 °C. Besides, the relative decrease in R_p that can be
162 theoretically ascribed to temperature-mediated variations in water viscosity (η) was also
163 calculated from the relative decrease in η in relation to its value at 12 °C. To this end, the
164 relationship between η and temperature reported by Roderick and Berry (2001) was used:

$$165 \quad \eta = 1.95 \times 10^8 / T^7 \quad (2)$$

166 where T is the liquid temperature in K and η is expressed in MPa s⁻¹.

167 *Statistics*

168 Statistical analyses were performed with Statistix (Statistix 10 for Windows, Analytical
169 Software, Tallahassee, FL, USA). Analyses of variance, with prior data transformation when
170 required, were used for comparing cultivars. Means were separated using the Tukey HSD test
171 when $P < 0.05$. When the assumptions of ANOVA could not be satisfied, the Kruskal-Wallis
172 test was used, being the distribution of the scores compared with the Dunn's test.

173

174 **Results**

175 *Experiment I*

176 The flow-pressure measurements conducted at 25 °C in Experiment I revealed average R_p
177 values ranging from $0.66 \cdot 10^4$ (in ‘Picual’) to $1.13 \cdot 10^4$ MPa m² s kg⁻¹ (in ‘Arbequina’) (Figure
178 2a). Albeit considerable, these differences were not statistically significant due to large
179 variability between replicates (average coefficient of variation of 32 %). On the other hand,
180 root surface in ‘Arbequina’ was significantly lower than in ‘Picual’, ‘Hojiblanca’ and
181 ‘Frantoio’ (Figure 2c).

182 The average cumulative distribution function of root diameters for each cultivar is shown in
183 Figure 3. In all cases, more than 50 % of the roots had diameters below 0.3 mm, while those
184 with diameters above 0.7 mm represented less than 10 %. The frequency distribution of root
185 diameters (Fig. S1) was unimodal, with roots between 0.2 to 0.3 mm in diameter being the
186 most frequent class. Apart from this, Figure 3 also reveals slight cultivar variability in the
187 frequency distributions leading to differences in the average root diameter. The latter ranged
188 from 0.27 to 0.36 mm for ‘Hojiblanca’ and ‘Arbequina’, respectively. Differences between
189 these two cultivars were significant (Figure 2e).

190 *Experiment II*

191 For all cultivars, pressure-flux measurements showed a progressive decrease in R_p as
192 temperature increased, changing from an average value of $3.5 \cdot 10^4$ MPa m² s kg⁻¹ at 12 °C to
193 $1.4 \cdot 10^4$ MPa m² s kg⁻¹ at 32 °C (Table 1). As in Experiment I, ‘Arbequina’ always exhibited
194 the highest R_p and ‘Picual’ showed the lowest values, except for the comparisons at 32 °C,
195 when the lowest values were estimated for ‘Hojiblanca’. Differences between cultivars were
196 not significant at 12 °C, but they became so at higher temperatures: R_p values in ‘Arbequina’
197 was significantly higher than those of ‘Picual’, ‘Hojiblanca’, ‘Arbosana’ and ‘Frantoio’ when
198 comparisons were conducted, respectively, at temperatures equal or higher than 16, 20, 20 and
199 32 °C. Differences between ‘Picual’, ‘Hojiblanca’, ‘Arbosana’ and ‘Frantoio’ were never

200 significant. As an additional interesting finding, R_p values at 25 °C in Experiment II were higher
201 than those determined in Experiment I, irrespective of the cultivar (Figure 2b).

202 During the course of the outdoor measurements, R_p decreased as temperature increased, and
203 cultivar differences were found in the extent of the relative R_p reduction (Figure 4). In this
204 regard, the increase in temperature from 12 to 32 °C yielded a 65–70 % reduction in R_p for
205 ‘Hojiblanca’ and ‘Arbosana’ and a 55–60 % reduction for ‘Arbequina’ and ‘Frantoio’.
206 Nonetheless, it is also noteworthy that there was considerable variability in the extent of the
207 relative decay of R_p with temperature between replicates in some cases (e.g. ‘Hojiblanca’).
208 Finally, Figure 4 also illustrates that viscosity effects are only able to explain a 38 % decrease
209 in R_p for the temperature interval considered.

210 Root systems in Experiment 2 presented higher A than those of Experiment 1, with the
211 comparison between cultivars leading to similar results (Figure 2d). The root diameter
212 frequency distributions in both experiments were virtually the same, regardless of the cultivar.
213 A non-significant trend to higher d was observed in Experiment 2 (Figure 2e-f).

214

215 **Discussion**

216 So far, reports on R_p are scarcely found within the scientific literature, let alone papers
217 exploring the genotypic variability in this trait. In fact, to the best of our knowledge, only four
218 past studies have analyzed differences in R_p between cultivars or rootstocks of a given species.
219 Aroca et al. (2001) compared the response of root hydraulic conductivity (i.e. the inverse of
220 R_p) after exposing plants of two maize genotypes to chilling temperatures. Statistical
221 differences between genotypes were significant at 5 °C, but not at a control temperature of 25
222 °C. In a similar study, Lee et al. (2005a) found significant differences in the hydraulic

223 conductivity of cucumber (*Cucumis sativus*) and figleaf gourd (*Cucurbita ficifolia*) roots, both
224 at low and control (25 °C) temperatures. By contrast, Bloom et al. (2004) found similar values
225 in the hydraulic conductance of excised roots when comparing cultivated tomato
226 (*Lycopersicon esculentum*) and a wild congener (*L. hirsutum*) irrespective of temperature.
227 Finally, García-Tejera et al. (2016) compared R_p for two common almond rootstocks (GF677
228 and GN15) at 25 °C, finding significant differences between them. In the present study, we
229 explored the genotypic variability in R_p between five widely used olive cultivars. We did not
230 find statistical differences between them when determinations were performed at two
231 contrasting steady temperatures (i.e. 25 °C in Experiment I and 12 °C in the first measurement
232 in Experiment II) despite some variability was observed in root traits (Fig. 2, Fig. 3). Partly,
233 the lack of significant differences was due to the large variability between replicates. This was
234 not an exclusive issue of our study, as similar levels of variability between replicates have also
235 been observed in previous reports (e.g. Cochard et al. 2000, Lee et al. 2005a). An explanation
236 for this phenomenon is still missing and clearly deserves further research.

237 Despite the variability in the studied species, plant material characteristics and methods, the
238 current body of literature consistently indicates that R_p is particularly sensitive to temperature
239 conditions, so that the lower the temperature in the root environment, the higher the R_p (Ramos
240 and Kaufmann 1979, Cochard et al. 2000, Aroca et al. 2012, García-Tejera et al. 2016). The
241 same trend was reproduced in Experiment II, irrespective of the cultivar (Table 1, Fig. 4).
242 Changes in sap viscosity drive the inverse relationship between R_p and temperature, but
243 frequently R_p variations cannot be entirely ascribed to that cause alone (Kuiper 1964, García-
244 Tejera et al. 2016). In fact, in our study decreases in R_p in the 12–32 °C interval were
245 considerably higher than the theoretical decrease in water viscosity (on average 17–32 %
246 higher, depending on the cultivar, Fig. 4). Modifications in cell membrane structure and
247 inhibition of aquaporin activity at low temperatures have been proposed as possible

248 explanations for the variations of R_p beyond the contribution of water viscosity (Wan et al.
249 2001, Aroca et al. 2005, Lee et al. 2005b, Murai-Hatano et al. 2008, Aroca et al. 2012).

250 Traditionally, temperature effects on R_p have been studied by determining R_p in different plants
251 (i.e. excised root systems) exposed to various temperatures (e.g. Aroca et al. 2001, Lee et al.
252 2005a, García-Tejera et al. 2016). This approach can generate some bias due to non-
253 homogeneity in root structural properties, as temperature comparisons are performed for
254 different plants. Alternatively, in some works, temperature comparisons have been made for
255 the same individuals by performing consecutive pressure-flow measurements changing
256 temperature in steps and waiting for the exudation rate to stabilize (Cochard et al. 2001, Wan
257 et al. 2001), with the only uncertainty arising from the fact that it implies working with a
258 detached root system for long periods. In Experiment II, we used a new approach aimed at
259 estimating short-term changes in R_p in response to a gradual continuous warming from 12 °C.
260 This allowed us to perform the pressure-flow measurements for the same root system under a
261 wide range of temperatures and within a relatively short period of time (5 h). Moreover, the
262 employed experimental setup is more suitable to mimic the rapid temperature variations to
263 which roots in the upper soil layers are exposed on a diurnal time scale (Villalobos et al. 2016).
264 Results from those measurements revealed two interesting findings. First, the estimates of R_p
265 at 25 °C were consistently higher than those obtained under steady conditions in Experiment I
266 (Figure 2a-b), which implies that it takes time for the non-viscous mechanisms inhibiting R_p at
267 low temperatures to deactivate completely after warming. This was not entirely unexpected, as
268 some reports indicate that it can take hours (Aroca et al. 2001, Lee et al. 2005a) or even days
269 (Wan et al. 1999) for R_p to reach an equilibrium value following a drastic temperature change.
270 In rice plants, this phenomenon has been linked to a coordinated up-regulation of root
271 aquaporin gene expression during the exposure to low temperatures (Ahamed et al. 2012).
272 Second, statistical cultivar differences were found in the short-term R_p responses to temperature

273 (Table 1), evidencing some genotypic variability in the inertia of the mechanisms controlling
274 membrane permeability at low temperatures. In other words, our results suggest that the time
275 required for olive roots to recover R_p back to initial values after a chilling period differs between
276 cultivars. Besides, it must be noted that, coming from warm field conditions, plants in
277 Experiment II were exposed to 12 °C for around 14 h before the start of pressure-flow
278 measurements. While this time might not have been sufficient for the plants to fully acclimate
279 in terms of R_p , cultivar differences at such temperature were not significant. This should
280 indicate that the differential response to temperature observed between cultivars is only
281 apparent at a very short-term scale (minutes or few hours).

282 A common effect of low (non-freezing) temperatures on plants is leaf dehydration (Aroca et
283 al. 2012). In olive trees and other sensitive species, low temperatures result in a decrease of
284 leaf water potential, even when soil water content is not limiting (López-Bernal et al. 2015,
285 Centeno et al. 2018). This chilling-induced dehydration is originated by an imbalance between
286 root water uptake and leaf transpiration, which is reportedly associated with the low-
287 temperature driven increase in R_p (Running and Reid 1980, Aroca et al. 2001, Centeno et al.
288 2018). Pérez-López et al. (2010) assessed the effects of soil chilling in the water relations of
289 six olive cultivars, among which ‘Picual’, ‘Arbequina’ and ‘Frantoio’ were included.
290 According to their results, the authors labelled the former two as ‘tolerant’ and the latter as
291 ‘sensitive’ to chilling-induced dehydration. Of these three cultivars, ‘Arbequina’ and ‘Picual’
292 always exhibited the highest and lowest R_p values in our experiments, respectively, although
293 the differences between them were not significant when measurements were performed at
294 steady temperatures. This is at odds with the fact that both were classified in the same category
295 (‘tolerant’), while the ‘sensitive’ cultivar ‘Frantoio’ showed intermediate R_p values. Therefore,
296 our findings suggest that there is no clear connection between the reported differences in the
297 sensitivity to soil chilling and a hypothetical variability in the R_p responses to temperature

298 between olive cultivars. However, we might speculate about a possible link between R_p
299 responses to temperature, chilling-induced dehydration and the winter dormant state of this
300 species.

301 In olive trees, vegetative growth ceases in autumn and undergoes a winter rest period lasting
302 until favourable conditions return in early spring. López-Bernal et al. (2020) performed a series
303 of experiments indicating that low temperatures control growth cessation and dormancy
304 induction in olive shoot apical meristems. According to the present study, the low temperatures
305 ($< 15\text{ }^\circ\text{C}$) leading to dormancy induction in autumn should also trigger an increase in R_p and
306 chilling-induced dehydration (Pérez-López et al. 2010). Given that growth processes are
307 inhibited at high water potentials (Hsiao 1973), autumn growth cessation might be associated
308 with a progressive increase in R_p and worsening of water status. On the other hand, it is
309 noteworthy that even if mean daily temperatures are low in winter, appropriate conditions for
310 growth can still occur around midday in many areas of the Mediterranean Basin. The absence
311 of olive vegetative growth during these periods might be related to the occurrence of high R_p
312 and low water potentials, as soil temperature should remain similar to the mean daily air
313 temperature (García-Tejera et al. 2016).

314

315 **Conclusions**

316 In conclusion, our findings show limited variability in R_p between the studied olive cultivars.
317 Nevertheless, the wide number of cultivars of different origins and the outcrossing nature of
318 this species (Díez et al. 2015) makes the existence of substantial genetic variability in this trait
319 still possible. As a consequence, further research with other cultivars would be needed to
320 elucidate whether it is possible to find genetic variability in R_p . With regard to the temperature
321 responses, it seems that, beyond the associated viscosity variations, the current study suggests

322 that additional mechanisms, such as changes in membrane permeability, may affect R_p , as
323 pointed out by other studies. These changes are not immediate, taking some time to reach a
324 ‘steady state’, which explains why our values of R_p at 25 °C varied between experiments. The
325 fact that R_p values became significantly different between cultivars with warming suggest that
326 the pace of temperature-induced changes in membrane permeability is cultivar-dependent. In
327 other words, some olive cultivars seem to have the capacity to acclimate R_p faster to rapid
328 temperature changes than others.

329

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341

342 **Conflict of interest**

343 None declared.

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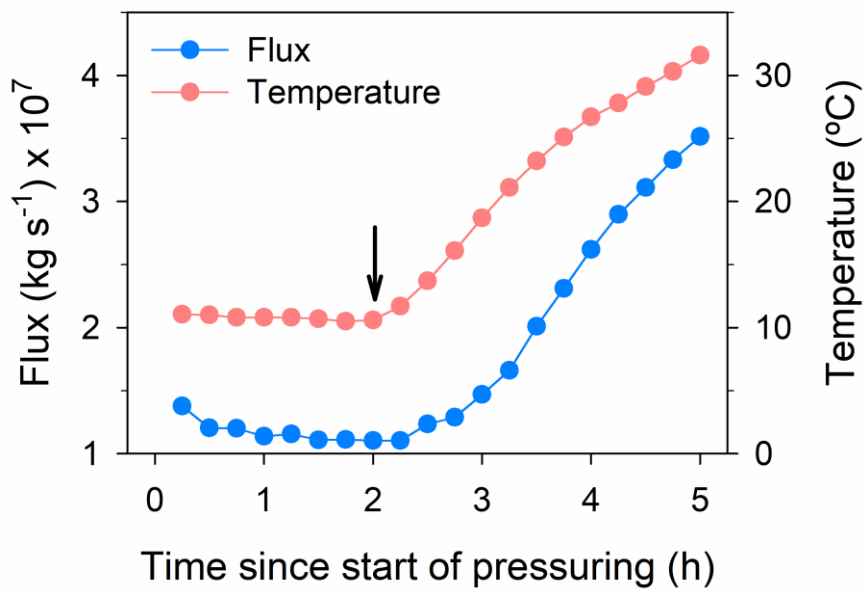
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449 **Figure captions**

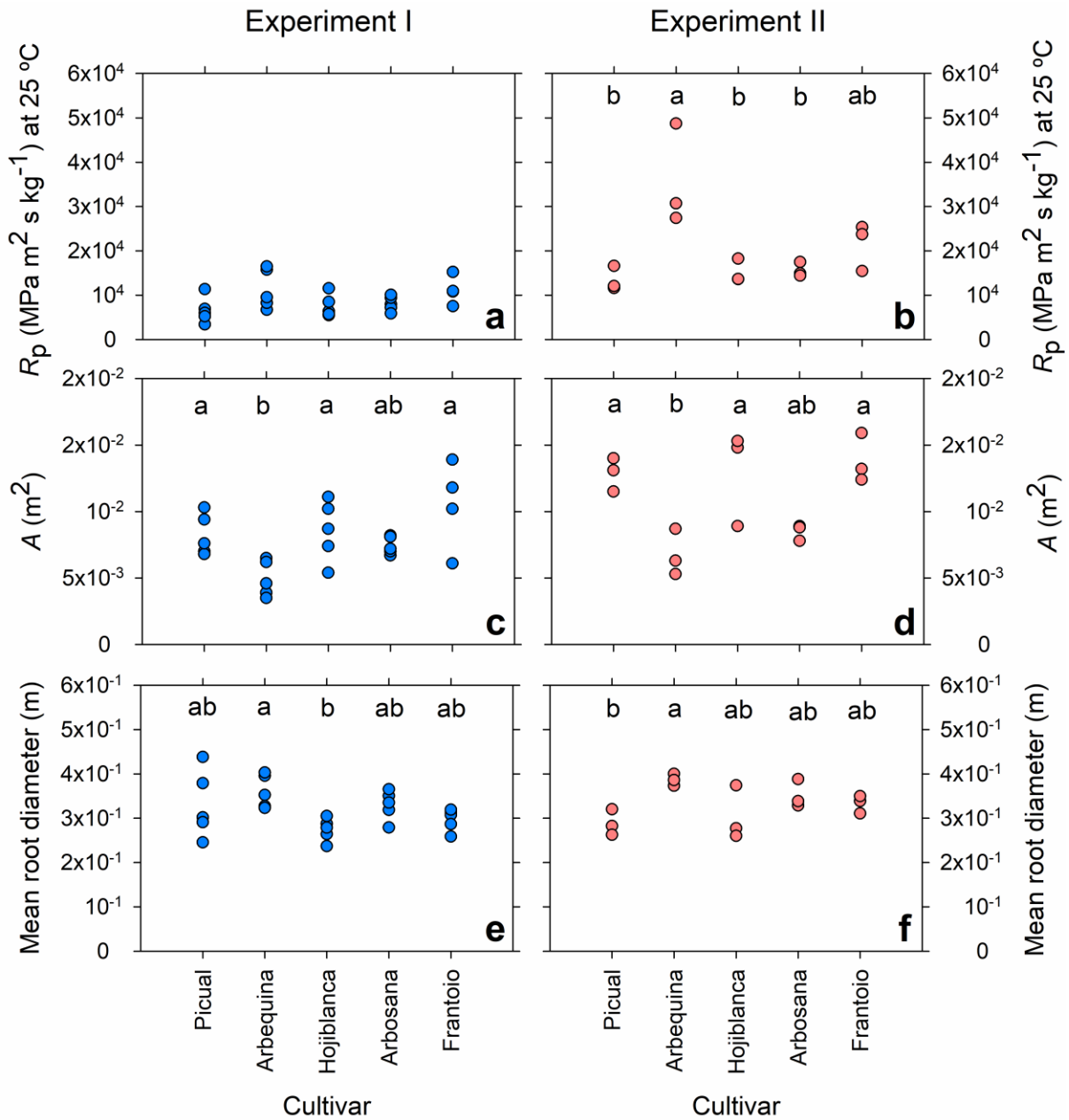
450 **Figure 1:** Time course of xylem flux and temperature (15-min averages) of the root system
451 over the measurement period of one of the ‘Arbosana’ replicates. The arrow marks the time at
452 which the equipment was transferred from the growth chamber to an outdoor sunlit location.



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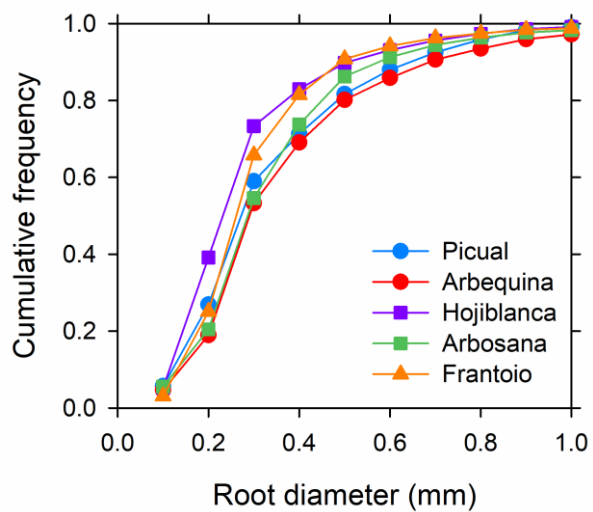
455 **Figure 2:** Measured values of (a, b) specific radial root resistance (R_p) at 25 °C, (c, d) total root
 456 surface (A) and (e, f) average root diameter for the different olive cultivars, both for Experiment
 457 I (n=5, left panels) and Experiment II (n=3, right panels). Each point corresponds to one of the
 458 replicates. Different letters denote significant differences ($P<0.05$) between cultivars.



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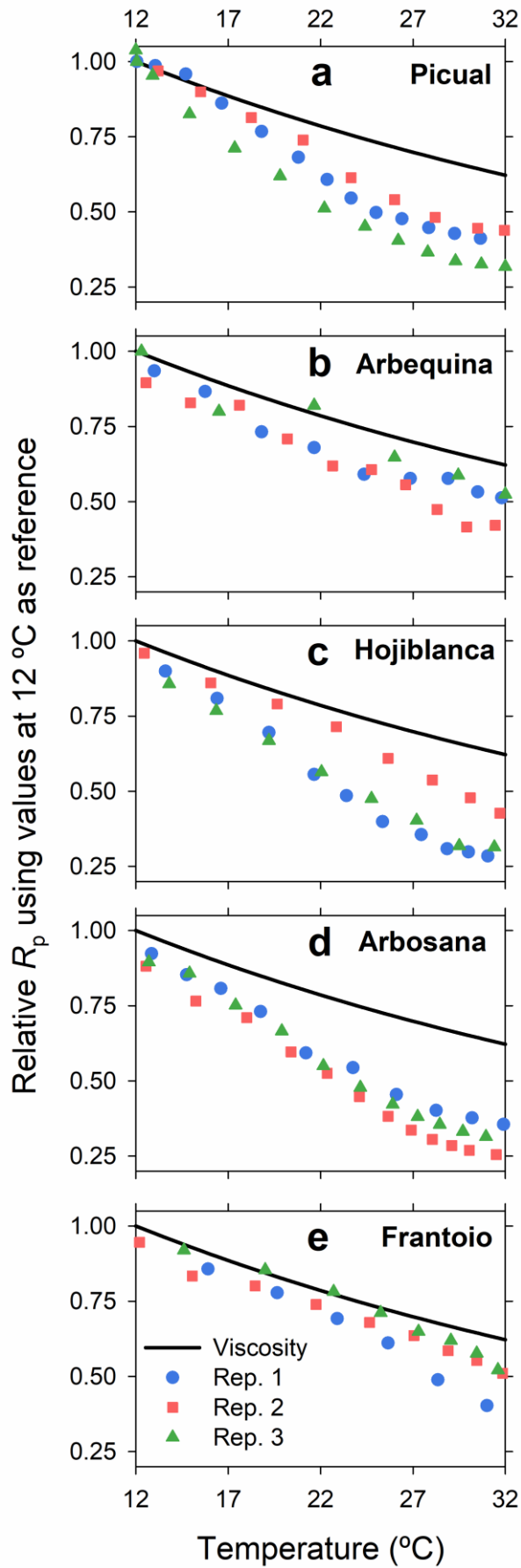
461 **Figure 3:** Cumulative distribution function of root diameters below 1.4 mm for the studied
462 olive cultivars in Experiment I.



463

464

465 **Figure 4:** Relative changes in specific radial root resistance (R_p) versus root temperature with
466 respect to the value estimated at 12 °C in Experiment II. Each panel corresponds to one of the
467 studied cultivars: (a) ‘Picual’, (b) ‘Arbequina’, (c) ‘Hojiblanca’, (d) ‘Arbosana’ and (e)
468 ‘Frantoio’. Symbols correspond to the three replicates per cultivar. The solid line represents
469 the relative variations in R_p that can be ascribed theoretically to temperature-mediated changes
470 in water viscosity.



472 **Tables**

473 **Table 1:** Specific radial root resistance (R_p , MPa m² s kg⁻¹ x 10⁴) at different temperatures for
 474 each of the olive cultivars in Experiment II. Values for each combination of temperature and
 475 cultivars correspond to the average of three individuals. Different letters denote significant
 476 differences (P<0.05) between cultivars.

477

Cultivar	Temperature (°C)					
	12	16	20	24	28	32
Picual	2.77	2.31 b	1.84 b	1.44 b	1.15 b	1.04 b
Arbequina	5.52	4.83 a	4.22 a	3.67 a	3.13 a	2.60 a
Hojiblanca	2.88	2.50 ab	2.06 b	1.62 b	1.25 b	1.00 b
Arbosana	3.24	2.74 ab	2.20 b	1.69 b	1.26 b	0.98 b
Frantoio	3.13	2.83 ab	2.51 ab	2.19 ab	1.86 ab	1.50 b

478