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Gualberto-Fernandes, T.J.; Campo García, ADD.; Herrera Fernandez, R.; Molina Herrera, A. (2016). Simultaneous assessment, through sap flow and stable isotopes, of water use efficiency (WUE) in thinned pines shows improvement in growth, tree-climate sensitivity and WUE, but not in WUEi. *Forest Ecology and Management*. 361:298-308.  
doi:10.1016/j.foreco.2015.11.029



The final publication is available at

<http://doi.org/10.1016/j.foreco.2015.11.029>

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Additional Information

1 **Simultaneous assessment, through sap flow and stable isotopes, of water use efficiency**  
2 **(WUE) in thinned pines shows improvement in growth, tree-climate sensitivity and**  
3 **WUE, but not in WUEi.**

4

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## 16 **Abstract**

17 In water-limited regions, adaptive management of forest and water relationships has been put  
18 forward, to implement hydrology-oriented silviculture to reduce stand evapotranspiration and,  
19 at the tree level, to improve growth and water use efficiency (WUE). The main goal of this  
20 study was to evaluate the effect of thinning in the short and medium term on tree growth,  
21 climate (drought) sensitivity, WUE performed using growth and sap flow measurements and  
22 WUEi performed using  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  isotopes, in a typical semiarid forest. This approach

23 also evaluated the reliability of isotopes as indicators of the effects of adaptive forest  
24 management. A stagnated Aleppo pine plantation was experimentally thinned at high intensity  
25 (H98) in 1998 and at High (H), Medium (M) and Low (L) intensities in 2008, along with a  
26 control (C). Substantial limitation of tree growth was observed in C. Thinning not only  
27 increased growth, but also changed the tree growth-precipitation relationships, with C trees  
28 depending more on precipitation than thinned trees did. WUE<sub>i</sub> after thinning was significantly  
29 affected only in the medium term, with C trees being more efficient (94.4  $\mu\text{molCO}_2/\text{molH}_2\text{O}$ )  
30 than H98 trees (88.7), especially in dry spells (100.7). WUE<sub>i</sub> was found to increase when  
31 precipitation decreased, regardless of the treatment. However, WUE increased sharply from C  
32 (1.26 g biomass/L H<sub>2</sub>O) to H (3.20 g/L), showing a clear difference with WUE<sub>i</sub> observed in  
33 the same years. Thinning caused an increase in  $\delta^{18}\text{O}$  in the short term, but no relationship was  
34 found between  $\delta^{18}\text{O}$  and tree water use. It can be concluded that forest management improved  
35 WUE in spite of higher tree transpiration, but WUE<sub>i</sub> remained unchanged, probably due to an  
36 underestimate of photosynthetic capacity. The dual isotope ( $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$ ) conceptual model  
37 was not consistent with our experimental data. Thus, the question of whether stable isotopes  
38 can be used as a tool for addressing the ecophysiological impacts of thinning remains open.

39 **Keywords:** Dendroclimatology; Adaptive forest management; Hydrology-oriented  
40 silviculture; *Pinus halepensis*; Aleppo pine; Tree transpiration.

41

## 42 **1. Introduction**

43 Water resource availability in the Mediterranean will be seriously jeopardized in the  
44 foreseeable future (García-Ruiz et al., 2011), which may have a substantial impact on the  
45 semiarid forests growing in the region (Lindner et al., 2010; Torras et al., 2012). To improve  
46 forest resilience, forest managers need adaptive strategies that help make forest management  
47 more efficient and effective under changing water resource availability (Sjölund and Jump,

48 2013). However, although many studies have addressed this need in recent years, reliable  
49 guidelines for adaptive management in line with the eco-regional and social context are still  
50 scarce (Fitzgerald et al., 2013).

51 In water-limited regions, adaptive management usually focuses on forest and water  
52 relationships based on hydrology-oriented silviculture (Del Campo et al., 2014; Ungar et al.,  
53 2013). Guidelines for this silviculture should be developed through a full approach covering  
54 tree to stand scales. Stand scale is related to the hydrologic performance of the physical  
55 structure of forests (density, LAI, canopy storage etc.), in which thinning is known to affect  
56 water cycle components such as rainfall interception, throughfall, transpiration, soil moisture  
57 and deep infiltration (Del Campo et al., 2014; Molina and Del Campo, 2012). On the other  
58 hand, tree-scale studies provide fundamental information about how changing forest structure  
59 and microclimate might lead to modifications in the ecophysiology of photosynthesis,  
60 transpiration and tree water relations (Aussenac, 2000). In this context, tree climate-growth  
61 relations, water use (WU) and water use efficiency (WUE) are central topics to be tackled  
62 when relating forest management and tree water (Brooks and Mitchell, 2011; Forrester et al.,  
63 2012; Kruse et al., 2012; Sohn et al., 2013; Ungar et al., 2013). While WU is the amount of  
64 water transpired by trees (Burgess et al., 2001), WUE is the ratio of carbon gain per WU  
65 (Brienen et al., 2011). In most of the literature, WUE is discussed either in terms of an  
66 instantaneous measurement of the efficiency of carbon gain per water loss, i.e. intrinsic water-  
67 use efficiency (WUE<sub>i</sub>), or as an integral of such efficiency over time, commonly expressed as  
68 the ratio of water use to biomass accumulation or to harvestable yield (Dye, 2000; Hubbard et  
69 al., 2010).

70 Plant carbon stable isotope composition ( $\delta^{13}\text{C}$ ) provides a time-integrated proxy of  
71 WUE<sub>i</sub> during the growing season (Dawson et al., 2002; Farquhar et al., 1989), because the  
72 isotopic carbon discrimination of plants is linearly linked to the  $C_i/C_a$  ratio, where  $C_i$  is the

73 partial pressure of CO<sub>2</sub> in the leaf intercellular spaces and C<sub>a</sub> that of the ambient air (Farquhar  
74 et al., 1982; Sheidegger et al., 2000). However, increases in δ<sup>13</sup>C, interpreted as a reduction in  
75 C<sub>i</sub>, may be the result of either i) reduced stomatal conductance (g<sub>s</sub>) at a constant  
76 photosynthetic capacity (A) or ii) increase in A at a constant g<sub>s</sub>, i.e. changes in WUE<sub>i</sub> are due  
77 to changes in transpiration rate or in photosynthesis activity. To overcome this, the dual-  
78 isotope model (Sheidegger et al., 2000) was developed to constrain the interpretation of δ<sup>13</sup>C  
79 variations in organic matter by measuring δ<sup>18</sup>O in the same material. In principle, the latter as  
80 a proxy for evaporative demand would be modified by g<sub>s</sub> but not by A, thus allowing for a  
81 better interpretation of WUE<sub>i</sub>. This model is conceptually sound and many authors have used  
82 it to interpret δ<sup>13</sup>C results measured in tree rings (Brooks and Mitchell, 2011). However,  
83 Roden and Siegwolf (2012), on analysing the systematic use of this conceptual model, warned  
84 about specific situations in which its applicability might be compromised.

85         Studies of WUE<sub>i</sub> and of the use of stable isotopes in Aleppo pine have been profuse in  
86 the literature. Previous studies used this technique to assess climate-growth relations (Ferrio  
87 et al., 2003), intra-specific differences in WUE<sub>i</sub> related to seed origin (Voltas et al., 2008) and  
88 differences in plantation performance (Querejeta et al., 2008), or to specifically address tree  
89 water use in the species (Klein et al., 2013a). Some of these studies provide key information  
90 for improving silviculture in the face of increasing water scarcity in Mediterranean regions.  
91 However, how forest management affects WUE, how long these changes may last and how  
92 stable isotopes can study these effects remain unclear for this species. For instance, Moreno-  
93 Gutiérrez et al. (2011, 2012) report no variation in WUE<sub>i</sub> in Aleppo pine after reducing forest  
94 density; similar results were found by Martín Benito et al. (2010) for black pine. In addition,  
95 Gyenge and Fernández (2014) report that thinning increased the amount of water reaching the  
96 soil, but that WUE<sub>i</sub> was unrelated to growth, nitrogen and light use efficiencies. On the other  
97 hand, Querejeta et al. (2008) report a significant effect of the afforestation method on WUE<sub>i</sub>

98 of Aleppo pine saplings. In addition, in most studies WUE is addressed by studying  $\delta^{13}\text{C}$  in  
99 tree-rings (i.e. WUEi), although some divergences may appear when  $\delta^{13}\text{C}$  results are  
100 compared to other techniques such as leaf-scale gas exchange (Klein et al., 2013a).

101 The present study complements our previously published stand-scale results from a  
102 stagnated Aleppo pine plantation (Del Campo et al., 2014) and specifically addresses: i) What  
103 are the short- and medium-term effects of thinning intensity on WUE and growth of trees? ii)  
104 Do these effects change in drought periods? iii) Is there congruency between WUEi findings  
105 with the isotope-based approach and WUE findings obtained from tree growth and tree water  
106 use by sap flow measurements? We also addressed the question of whether the dual isotope  
107 technique ( $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$ ) could be used to explain further the WUE results.

## 108 **2. Material and Methods**

### 109 **2.1. Experimental site and design**

110 The experimental site and design have been described elsewhere (Del Campo et al.,  
111 2014). Briefly, “La Hunde” site is located in Valencia, Spain (39°5′N; 1°13′W, 943 m a.s.l.).  
112 The climate is Mediterranean with an average total annual precipitation of 477 mm and a  
113 mean annual temperature of 14.1°C. The soils are shallow (50-60cm) with a sandy-silty  
114 texture and basic pH. The area is occupied by *Pinus halepensis* Mill. plantations about 50-60  
115 years old and with high tree density (ca. 1,500 trees/ha) due mainly to low forest  
116 management. The experimental set-up encompassed five treatments. One of them was  
117 selected within a non-replicated plot heavily thinned in 1998 (H98), leaving approximately  
118 10% of the trees. This 0.2 ha plot was established and sampled to assess the medium-term  
119 effects of thinning. Adjacent to this plot, another experimental area was set up. This consisted  
120 of a randomized block design with three blocks (0.36 ha each) to assess the short-term effects  
121 of thinning. Each block was further divided into four plots (30x30m), three of them with

122 thinning treatments performed in 2008 at different intensities (High-H, Medium-M and Low-  
123 L) and a control plot (C) common to both experimental areas (Table 1).

## 124 **2.2. Tree growth**

125       Between eight and twelve trees per treatment were cored (north and south) by a 5mm  
126 increment borer at 1.30 m height. Each core was mounted on a wooden support and sanded  
127 until wood cells were clearly identified under the stereomicroscope. All cores were visually  
128 cross-dated and measured to the nearest 0.01mm with a measuring table (LINTAB 6.0, Frank  
129 Rinn, Heidelberg, Germany) coupled with the TSAP-Win software package (Rinn, 2011).  
130 Cross-dating of the tree-ring width (TRW) series was evaluated by the COFECHA software  
131 (Holmes, 1983). Cores with missing rings were ruled out for any further analysis. The average  
132 series length was 50.2 ( $\sigma=2.51$ ) years; autocorrelation at 1-year average was 0.76 ( $\sigma=0.09$ );  
133 and the Gini coefficient, which describes annual changes in the inequality of size and size  
134 increment, was 0.42 ( $\sigma= 0.08$ ), where 0 indicates perfect equality (the size or growth of all  
135 individuals is the same) and 1 indicates perfect inequality. The average series correlation to  
136 the master chronology was 0.79 ( $\sigma=0.07$ ), excluding the H98 treatment, which was analysed  
137 separately, obtaining a correlation of 0.80 ( $\sigma=0.09$ ).

138       The tree ring width (TRW) series were detrended to reduce the systematic noise  
139 caused by tree age (Cook and Briffa, 1990), using a cubic smoothing spline function with a  
140 wavelength fixed at 67% (Cook et al., 1990) of the length of the series, and a 50% frequency  
141 response. In some cores a negative exponential method was used instead. Each measured  
142 series was standardized by dividing the observed values by the predicted ones to obtain  
143 dimensionless TRW index series (TRWi). TRWi was averaged using a robust bi-weight  
144 mean. Additionally, the temporal autocorrelation was removed from each series by an  
145 autoregressive model (Cook and Briffa, 1990) to obtain the standard and residual chronology.

146 To determine the length of the residual chronology for which climatological responses would  
147 be tested, a running (20-year) mean of the expressed population signal (EPS) statistic was  
148 used to provide an indication of chronology reliability (Wigley et al., 1984).

149 Then, TRW data were used to estimate annual biomass increment value by an  
150 allometric equation developed for *Pinus halepensis* Mill. in the Mediterranean area (Gracia et  
151 al., 2004). Factors that might affect allometric relationships in this species vary with stand  
152 density, age, site and recent weather conditions (López-Serrano et al., 2005). Because there is  
153 no reported study on this species with a similar combination of these factors, we decided to  
154 opt for allometric models based on regional forest inventory data (Gracia et al., 2004), which  
155 cover a wide ecological range and a good representation of Aleppo pine forest conditions and  
156 trends across the landscape. The allometric equations used come from pure or mixed stands  
157 (basal area of *Pinus halepensis*  $\geq 70\%$ ) with a minimum canopy cover of 40%. The  
158 equations allow total aboveground biomass to be calculated (N=2,206 trees,  $R^2=0.61$ ,  
159 standard error of estimation 0.74 kg). Belowground biomass was assumed to be 0.30 of the  
160 aboveground value, in the range 0.23-0.35 previously reported for this species (Alfaro-  
161 Sánchez et al., 2015 and references therein).

### 162 **2.3. Water use and meteorological determinations**

163 Sap flow was measured by the heat ratio method (HRM) (Burgess et al., 2001) in four  
164 trees per treatment from April 2009 to May 2011, as described in Del Campo et al. (2014).  
165 The years 2009 and 2010 from these records were used in this study to calculate water use  
166 efficiency (WUE). An artificial neural network model (Fernandes et al., 2015) estimated  
167 water use in the months when data were missing or absent. The trees in which sap flow was  
168 measured were cored for the growth and isotope analyses. WUE in each tree was calculated  
169 for the years 2009 and 2010, as the ratio between annual biomass increment (grams of dry



170 weight) and tree water use (litres). Daily data on precipitation and temperature were also  
171 available at the site. The long-term series (1960-2008) were obtained from the meteorological  
172 stations nearest to the study site (Ayora-La Hunde from SAIH-CHJ network and Almansa  
173 from SIAR network).

#### 174 **2.4 Carbon and Oxygen isotope analysis**

175 Stable isotopes in the growth rings for the 1995 to 2010 period for the C and H98  
176 treatments were analysed in order to explore the effect of thinning in the medium term. For  
177 the treatments thinned in 2008, the period analysed ran from 2004 to 2010. The samples for  
178 this isotopic analysis were taken in April 2012 from cores that had been previously measured  
179 and dated. The sanded core surfaces were thoroughly cleaned with ethyl alcohol p.a. and  
180 small sections, including both early and late wood, were taken with a surgical blade under the  
181 stereomicroscope. We sampled whole wood with no previous treatment to separate fractions,  
182 in order to minimize analysis time and resources. Recent literature has confirmed that, in  
183 Aleppo pine, whole wood provides more consistent and coherent relationships with climatic  
184 variables (Del Castillo et al., 2015; Klein et al., 2013b). Samples were ground in an agate  
185 mortar and weighed to the nearest 0.001 g in Ag capsules for  $\delta^{18}\text{O}$  analysis and in Sn capsules  
186 for  $\delta^{13}\text{C}$  analysis.

187 Isotopic signatures of the wood samples were measured on-line by an isotope ratio  
188 mass spectrometer (Finnigan MAT, delta S, delta XL plus, delta XP) *via* a Conflo II interface  
189 for the combustion/pyrolysis of organic material, at the Paul Scherrer Institut, Ecosystem  
190 Fluxes Research Group, Villigen, Switzerland. Stable isotope data are presented as  $\delta^{13}\text{C}$  and  
191  $\delta^{18}\text{O}$ , respectively, relative to the international VPDB standard (‰).

#### 192 **2.5 Intrinsic water use efficiency**

193 The isotopic discrimination ( $\Delta$  - Farquhar and Richards, 1984) between carbon from  
 194 atmospheric CO<sub>2</sub> and plant carbon from C<sub>3</sub> plants, resulting from the preferential use of <sup>12</sup>C  
 195 over <sup>13</sup>C during photosynthesis, is defined in Eq. 1.

$$196 \quad \Delta = (\delta^{13}C_{\text{atm}} - \delta^{13}C_{\text{wood}})/(1 + \delta^{13}C_{\text{wood}}/1000) \quad (1)$$

197 where  $\delta^{13}C_{\text{atm}}$  and  $\delta^{13}C_{\text{wood}}$  are the isotopic ratios of carbon (<sup>13</sup>C/<sup>12</sup>C) in atmospheric CO<sub>2</sub> and  
 198 plant material, respectively.

199  $\Delta$  is linearly related to C<sub>i</sub>/C<sub>a</sub>, which is the ratio of intercellular (C<sub>i</sub>) to atmospheric  
 200 (C<sub>a</sub>) CO<sub>2</sub> mole fractions, by Eq. 2 (Farquhar et al., 1982):

$$201 \quad \Delta = a + (b - a)C_i/C_a \quad (2)$$

202 where  $a$  is the fractionation during CO<sub>2</sub> diffusion through the stomata (4.4‰; O’Leary, 1981)  
 203 and  $b$  is the fractionation associated with reactions by Rubisco and PEP carboxylase (27‰;  
 204 Farquhar and Richards, 1984). C<sub>a</sub> and  $\delta^{13}C_{\text{atm}}$  were obtained for each year, using the relations  
 205 found by Feng (1999), by means of equations 3 and 4, respectively.

$$206 \quad C_a = 277.78 + 1.35 \times e^{0.01572 \times (\text{year} - 1740)} \quad (3)$$

$$207 \quad \delta^{13}C_{\text{atm}} = -6.429 - 0.0060 \times e^{0.0217 \times (\text{year} - 1740)} \quad (4)$$

208 We used this additional information, as described in Kruse et al. (2012), to estimate C<sub>i</sub>  
 209 and intrinsic water use efficiency (WUE<sub>i</sub>) for particular years, as affected by stand  
 210 development and plantation management, following thinning.

$$211 \quad C_i = \delta^{13}C_{\text{atm}} - \delta^{13}C_{\text{wood}} - a/b - a \quad (5)$$

$$212 \quad WUE_i = A/g = (C_a/1.6) \times (b - \delta^{13}C_{\text{atm}} + \delta^{13}C_{\text{wood}})/(b - a) \quad (6)$$

213 WUE<sub>i</sub> is usually expressed in  $\mu\text{molCO}_2/\text{molH}_2\text{O}$ .

## 214 2.6. - Data treatment and analysis

215 Data of TRW,  $\delta^{18}\text{O}$  and WUE<sub>i</sub> were tested for differences between the pre-treatment  
216 period (1995-1998 in H98, 2004-2007 in H, M and L treatments) and the post-treatment  
217 periods (2008-2010). In all cases the control (C) was also included. Before the analyses, data  
218 were tested for normality and variance homogeneity by the Shapiro-Wilk test and Levene  
219 tests, respectively. The t-student, ANOVA and Kruskal-Wallis tests were the procedures used  
220 for mean comparisons, using the R-package Agricolae (Mendiburu, 2014) and at p-level  
221 <0.05.

222 For the pair H98 and C, these analyses were also conducted in contrasting years by  
223 classifying them as dry or wet years. For this purpose, the Standardized Precipitation Index  
224 (SPI) (McKee et al., 1993) was calculated by the R-package SPEI index (Beguería and  
225 Vicente-Serrano, 2013) to adjust the precipitation series to a given Pearson III distribution  
226 probability (Pasho et al., 2011). Positive SPI values indicate greater than median  
227 precipitation; and negative values, lower than median precipitation. For the purpose of this  
228 study we considered that a drought event started when the SPI value reached -1.0 and ended  
229 when the SPI became positive again. A year was considered dry when the sum of the SPI for  
230 a 3-month time scale was less than zero; and wet, when it was greater than zero.

231 For the dendroclimatic analysis (influence of thinning on the dendroclimatic  
232 responses), the residual chronologies (TRWi) from Control and High 98 treatments were  
233 examined. The relationship between the residual chronologies and climate data in the time  
234 interval 1960-2010 was determined by bootstrapped response functions (Guiot, 1991). To  
235 investigate the stability of growth-climate relationships, the moving response by TreeClim R-  
236 package was used (Zang and Biondi, 2013, 2015). The coefficients of the response functions  
237 were obtained through principal-component regression (Fritts, 1976). Correlation analysis  
238 used Pearson's product moment correlation. The significance of correlation and response  
239 function coefficients was tested at 0.05 level using 1,000 bootstrapped estimates, obtained at

240 random by replacement from the initial data set (Wang et al., 2012; Zang and Biondi, 2015).  
241 The temporal window used for calculating growth-climate relations extends from April of the  
242 previous year to October of the growth year for the common period (Magruder et al., 2013),  
243 which was considered adequate to explain tree-ring width variations. All statistical  
244 procedures, except a cross-dating check, were performed using R 3.1.3 GUI 1.62 (R Core  
245 Team, 2015).

### 246 **3. Results**

#### 247 **3.1. Influence of thinning in the short and medium term**

248 Tree growth, expressed as TRW, varied a lot in the initial growth of the stand (ca. 15  
249 years), probably related to genetics and environmental conditions. This stage was followed by  
250 a steady decrease in both magnitude and variability, probably related to increased competition  
251 between trees (Fig. 1). In the following years, TRW values were very small, but still showed  
252 responses to climate conditions, especially during dry years (e.g. 1987-1988, 1994-1995 and  
253 2004-2005). Following thinning, the remaining trees showed significant increases in TRW  
254 compared with control ones (Fig. 1a, b).

255 Expressed population signals (EPS) of all TRW residual chronologies were above  
256 the critical value of 0.85 (Wigley et al., 1984). In addition, the mean of the correlations  
257 between series from each tree over all trees also indicated good agreement ( $>0.80$ ). In all  
258 chronologies there were TRW index reductions in dry years, whereas periods characterized by  
259 a substantial increase in radial growth were observed in wet years and after thinning at  
260 different scales (Fig. 1c, d). Despite the differences observed after thinning, no significant  
261 differences were found in the TRW index before and after thinning (data not shown).

262 Table 2 summarizes the differences for mean TRW in selected years before and after  
263 thinning. There were significant differences after thinning in both the medium-term (between

264 C and H98) and the short-term analysis, although in this case the lowest-intensity thinning (L)  
265 did not differ in growth from the Control.

266 Values for WUEi ranged from 80.6 to 114.9  $\mu\text{molCO}_2/\text{molH}_2\text{O}$  (Table 2), with  
267 significant differences after thinning between H98 and C. Values were higher in the Control  
268 trees, although this trend was not always consistent over time (Fig. 2a). In contrast, no  
269 significant difference was observed in the short-term analysis between any of the thinned  
270 treatments and the control treatment (Table 2), with only a marginal difference ( $p < 0.1$ )  
271 between H and C in the single-year comparison (Fig. 2b).

272 Values for  $\delta^{18}\text{O}$  ranged from 26.4 to 32.1‰, being very similar in all individual trees  
273 before thinning (average =  $28.5 \pm 0.84$ ‰), and increased after thinning in the short term (2008-  
274 2010) (Table 2), with significant differences between H and M treatments relative to C (Fig.  
275 3b-d). In the case of the H98 trees, the  $\delta^{18}\text{O}$  values (Fig. 3a) were on average 1.8‰ higher  
276 than those of the C trees from 1998 to 2004, but this difference dropped in the period 2005-  
277 2009 to only 0.3‰ and became non-significant in the 2008-2010 period (Table 2). These  
278 results indicate a short-term effect of thinning on  $\delta^{18}\text{O}$  that then drops in the medium term,  
279 although splitting the sample into wet/dry years yielded significant differences (see below).

### 280 **3.2. Relationships between thinning and climate**

281 Growth, WUEi and  $\delta^{18}\text{O}$  were also compared between H98 and C trees in specific  
282 dry and wet years according to the SPI index (Fig. 1S - supplementary material). In this  
283 analysis, significant effects of thinning on the three variables in the drier years and on TRW  
284 and  $\delta^{18}\text{O}$  in the wetter years were found (Table 3), with higher growth and  $\delta^{18}\text{O}$  in H98 and  
285 higher WUEi in C. The other treatments and Control could not be compared because there  
286 were not enough wet and dry years before and after thinning. However, there were different  
287 slopes on the response of WUEi to annual precipitation after thinning ( $F=11.03$ ;  $p=0.021$ ) and

288 no significant difference in the slopes of this response before thinning ( $F=0.27$ ,  $p=0.85$ ).  
289 Taking all treatments together,  $WUE_i$  increased significantly for dry years, regardless of the  
290 thinning treatment (Fig. 4).

291 The static bootstrapped correlation function between tree-ring residual chronologies  
292 (TRWi) and climate data (precipitation and temperature) showed different patterns between C  
293 and H98 plots, with more uniformity in the sign of correlations in C than in H98: a negative  
294 relationship with monthly temperature and a positive one with monthly precipitation (Fig. 2S  
295 supplementary material). By seasons, TRWi in C correlated with summer precipitation (June  
296 to August;  $r=0.50$ ) and correlated negatively with summer mean air temperature ( $r=-0.44$ ),  
297 both in the current year. For H98, only autumn temperature correlated with TRWi (September  
298 to November;  $r=0.37$ ) and no significant correlation with precipitation was found.

299 As growth-climate relationships are not necessarily stable over time, a bootstrapped  
300 moving-response function analysis was also performed (Fig. 5). In this case, a similar pattern  
301 between monthly precipitation and growth to that described above was observed: higher and  
302 positive dependence of growth on precipitation in C trees, which increases over time. That is  
303 to say, the H98 plot showed less consistent patterns between precipitation in the current year  
304 and growth (only a clear response with May precipitation, Fig. 5b) than the C plot (Fig. 5a),  
305 which showed a higher positive response with July to September precipitation of the current  
306 year. This pattern was especially clear during the 15 years after the thinning (Fig. 5): temporal  
307 windows from 1998 onwards show that the relationship between growth and monthly  
308 precipitation in the thinned plot varies between positive and negative, i.e. without a clear  
309 pattern (significant correlations were found only in April-May in this period); whereas in the  
310 Control the frequency of positive and significant correlations between growth and  
311 precipitation, whenever it occurs, is much higher.

### 312 3.3 WU, WUE, WUEi and $\delta^{18}\text{O}$

313 Water use, the mean annual transpiration per tree, was greatest in the trees from the  
314 H plot, followed by H98>M>L>C, although there was no statistical significance between C, L  
315 and M (Fig. 6). On average (years 2009 and 2010), a tree from high-intensity thinning (H)  
316 transpired more than three times the amount of water transpired by a control tree. However,  
317 water use efficiency (WUE), obtained from both these data and those from the annual  
318 increment of biomass in dry weight, showed that trees from both H- and M-intensity thinning  
319 were more efficient than the C trees (Fig. 6). The other thinning treatments (L and H98)  
320 showed gradually higher WUE than the C one did, but without significant differences (C:1.26,  
321 L:1.84, M:2.97, H:3.20 and H98:2.51 g biomass/L water transpired).

322 These data on WUE did not show any correlation with WUEi, which showed no  
323 differences between treatments (except in drier years in the medium-term analysis). In fact,  
324 there was a clear trend for the C trees to have higher values of WUEi in the years 2009 and  
325 2010 (Fig. 2). The units of WUEi,  $\mu\text{mol C fixed per mol H}_2\text{O}$ , are used as a relative indicator  
326 rather than an absolute value. However, it was thought useful to compare WUE and WUEi for  
327 the same trees and the same years and thus show the effect of treatments.

### 328 4. Discussion

329 Hydrology-oriented silviculture might assist in adapting semiarid forests to climate  
330 changes. However, to achieve this goal requires better understanding and quantification of  
331 tree-water relations and in particular of tree water use efficiency. Here, the effects of thinning  
332 in the short and medium term were studied by focusing on water use efficiency and climate-  
333 growth relationships in a semiarid, planted pine forest.

#### 334 **4.1. Influence of thinning in the short and medium term**

335 Thinning induced differences in tree growth rates, as expected (Del Campo et al.,  
336 2014). The results on the isotopes showed a medium-term effect of thinning on WUE<sub>i</sub> (higher  
337 for C trees) and a short-term effect on  $\delta^{18}\text{O}$  (lower for C trees). Similar findings were reported  
338 by Brooks and Mitchell (2011), with a significant increase in  $\delta^{18}\text{O}$  up to 6 years after thinning  
339 (thinning removed 50% of basal area), i.e. in the short term, and with little or no response on  
340 WUE<sub>i</sub> in both the long and the short term. Independence of WUE<sub>i</sub> on stand density reduction  
341 in the short term was reported for Aleppo pine by Moreno-Gutiérrez et al. (2011). These  
342 results suggest that medium to high thinning does not improve WUE<sub>i</sub> in spite of the important  
343 physiological changes induced in the remaining trees, as seen for growth. This may be  
344 because, under our experimental conditions, the years after thinning were very similar in  
345 terms of overall precipitation, despite the differences in net rainfall between plots (Del Campo  
346 et al., 2014) and the different seasonal distribution of precipitation (Fig. 3S and 4S -  
347 Supplementary materials). This is supported by the results found for dry years (Table 3) and  
348 the significant relation between WUE<sub>i</sub> and precipitation (Fig. 4). Strong correlation between  
349 WUE<sub>i</sub>,  $\delta^{18}\text{O}$  and growth with inter-annual changes in water availability is known in *Pinus*  
350 *halepensis* (Moreno-Gutiérrez et al., 2012).

351 Differences in  $\delta^{18}\text{O}$  were also enhanced in dry years in our study. However, our  $\delta^{18}\text{O}$   
352 results do not corroborate those reported by Moreno-Gutiérrez et al. (2011), in which the  
353 remaining Aleppo pine trees in the heavily thinned treatment had lower leaf  $\delta^{18}\text{O}$  than the  
354 moderately thinned ones. These authors attribute this to higher stomatal conductance due to  
355 decreased inter-tree competition for water, which is supported by Farquhar et al. (2007). In  
356 contrast, other authors (Martín-Benito et al., 2010) reported that changes in canopy  
357 microclimate after thinning (decrease in relative humidity and increase in leaf temperature)  
358 are the likely drivers for  $\delta^{18}\text{O}$  cell increases, which is also supported by Farquhar et al.



359 (2007). Our results corroborate this latter theory, suggesting that increases in  $\delta^{18}\text{O}$  might be  
360 due to the hotter and drier environment induced in the stand after thinning, but that this  
361 increase might not be related to changes in stomatal conductance.

#### 362 **4.2. Climate growth relationships after thinning**

363 Results have demonstrated that thinning makes trees less sensitive to water  
364 shortages, indicating that trees in the C plot need to rely more heavily on current year  
365 precipitation than those in H98. These results are common for several species (Martín-Benito  
366 et al., 2010; Simonin et al., 2007; Sohn et al., 2013; Zhang et al., 2013), although no data have  
367 been reported specifically for Aleppo pine, to the author's knowledge. Other authors have  
368 analysed climate-growth relations for this species (De Luis et al., 2011; Moreno-Gutiérrez et  
369 al., 2011, 2012; Pasho et al., 2012), but no management effect was assessed. As such, our  
370 main finding is the observed temporal trend of growth-climate relationship, varying over the  
371 trees' lifetime (Herrero et al., 2013; Magruder et al., 2013), especially after thinning. The  
372 dynamics of cambial activity in Aleppo pine is characterized by two major growth phases,  
373 one in spring (March-May) and another in autumn (September-November), interrupted in the  
374 summer period (De Luis et al., 2011), which in turn also affects tree-ring growth (Moreno-  
375 Gutiérrez et al., 2012). In our study, tree growth in the thinned plot (H98) showed a positive  
376 association with the previous summer's temperature and with May precipitation in the same  
377 year, thus taking advantage of the rainfall increase recorded in May during the last 15 years  
378 (Fig. 4S - Supplementary materials). On the other hand, the C plot was constrained by water  
379 deficit (Del Campo et al., 2014): any precipitation in the growing season fostered growth,  
380 with its relationship becoming significantly more positive as time passed and competition  
381 among trees increased. However, these relationships were lower in H98 as a consequence of  
382 less competition due to thinning, emphasizing the importance of water supply when the  
383 vessels are formed (González and Eckstein, 2003).

384           As these results confirm that thinning increases the resilience of Aleppo pine trees to  
385 climate variations, it should be seen as an effective use of adaptive silviculture to face climate  
386 changes. However, it seems that when thinning is delayed, the results might be less effective  
387 due to more prolonged stagnation, as observed in our contrasting experimental plots (H vs  
388 H98, Fig. 2).

### 389 **4.3. Interpreting trends in WUE, WUEi and $\delta^{18}\text{O}$**

390           Our WUE results for 2009 and 2010, estimated from water use through sap flow and  
391 biomass increment measurements, indicated that all thinning treatments improved the way  
392 trees used the soil water reservoir, but to different extents. WUE grew proportionally with  
393 decreasing forest cover, as indicated by a strong increase from C to M trees and similar values  
394 for M and H trees. Moreover, a high-intensity thinning treatment (H98) also showed (though  
395 non-significant) improvement in WUE 10 years after the intervention (ca. two-fold compared  
396 with C trees), but progressively decreased when compared to H. As the biomass term in the  
397 estimation of WUE is derived from diameter-based allometric equations, we suspect that  
398 some of our treatments (especially those with low densities) are out of the range covered by  
399 the equations. This is why we used up to six allometric equations from different authors  
400 (Jenkins et al. 2003; Grünzweig et al., 2007; Alfaro-Sánchez et al., 2015 and references  
401 therein), including models derived from low-density stands. Estimates of total tree biomass  
402 indicated that variations between treatments in our equations were similar and proportional  
403 within the different models (data not shown). In a nearby region with similar site, same  
404 species and different thinning treatments, Alfaro-Sánchez et al. (2015) reported that root  
405 growth was comparatively lower (root/shoot ratio) under high density and no management. If  
406 this occurred in our study, it would enhance the differences between treatments.

407           In terms of hydrology-oriented silviculture, the most interesting result was found  
408 when comparing tree water use, which clearly increased from C to H trees, to tree WUE,

409 which had an opposite trend. This indicates that trees in the M treatment, with a moderate soil  
410 water uptake between the C, L trees and the H trees, offset their water use by high growth  
411 rates, probably due to changes in A (photosynthetic rate), as may be expected from this forest  
412 structure reduction. Hale (2003) found that light environment below canopy was  
413 exponentially modified by basal area reduction through thinning in a Sitka spruce plantation.  
414 Therefore, and considering the hydrological data for these trees at the stand scale (Del Campo  
415 et al., 2014), we hypothesize that the opening created by the M treatment represents the initial  
416 stage in which substantial modifications in microclimatic conditions are produced, affecting  
417 leaf photosynthesis by improving light intensity and leaf temperature (Aussenac, 2000).  
418 Moreover, through the H, H98 and C comparisons, we observed that WUE was still higher 10  
419 years after thinning (H98 *versus* C), but less than the short-term increase (H98 *versus* H). This  
420 result indicates that the efficiency of silvicultural interventions on tree status may change over  
421 time (Aussenac, 2000), as could be expected, because of increased forest cover due to crown  
422 growth.

423 In contrast, the results from stable isotopes showed a non-significant effect of  
424 thinning on WUE<sub>i</sub> in the short term. Since WUE<sub>i</sub> estimated from  $\delta^{13}\text{C}$  may change due to  
425 alterations either in A (photosynthetic rate) or  $g_s$  (stomatal conductance) or both,  $\delta^{18}\text{O}$  was  
426 complementarily used as an indicator of  $g_s$  (Scheidegger et al., 2000). These authors  
427 described that  $\delta^{18}\text{O}$  in plant tissues may reflect three different sources of variation, i)  $\delta^{18}\text{O}$  in  
428 the source of water, ii) leaf water enrichment and iii) biochemical fractionation during the  
429 synthesis of organic matter. In this respect, several thinning studies have considered the  $\delta^{18}\text{O}$   
430 in the source of water and the biochemical fractionation during the synthesis of organic matter  
431 to be the same in control and thinned trees, when trees are collocated and are from the same  
432 species (Brooks and Mitchell, 2011; Martín-Benito et al., 2010). Therefore,  $g_s$  changes are  
433 directly related to the leaf water enrichment led by thinning. This would suggest that our

434  $\delta^{18}\text{O}$ -enriched treatments (H, M and H98) showed higher  $g_s$  and consequently higher water  
435 use, which is consistent with our experimental data on sapflow. However, other authors have  
436 pointed out that it is difficult to correlate  $\delta^{18}\text{O}$  and  $g_s$  (Barnard et al., 2012), since the  
437 environmental factors affecting leaf water enrichment should be constant among the sampled  
438 trees (Roden and Siegwolf, 2012). Our thinning treatments increased considerably the amount  
439 of water reaching the soil surface by reducing rainfall interception, but also increased  
440 evaporation from the soil (Del Campo et al., 2014; Molina and Del Campo, 2012). Therefore,  
441 we hypothesize that the assumption of constant  $\delta^{18}\text{O}$  in the soil water between the sampled  
442 trees may be violated by thinning causing evaporation to increase during rainfall and by  
443 higher direct evaporation from the soil surface. However, Moreno-Gutiérrez et al. (2011)  
444 found no differences in  $\delta^{18}\text{O}$  in xylem water (proxy for soil water) between moderate and  
445 heavy thinning treatments under similar experimental conditions.

446 To address the discrepancies we found between WUE and WUE<sub>i</sub>, we posited no  
447 differences in our source of water (as reported by Moreno-Gutiérrez et al., 2011) and thus  
448 assumed higher  $g_s$  in our  $\delta^{18}\text{O}$ -enriched trees (which was consistent with the sapflow results).  
449 Then, the lack of changes in WUE<sub>i</sub> as derived from  $\delta^{13}\text{C}$  might be due to an underestimation  
450 of A (photosynthetic rate), which must have increased proportionally more than  $g_s$  in order to  
451 agree with the WUE observed from biomass growth. Therefore, the question concerning the  
452 use of stable isotopes as a tool for determining the physiological impact of thinning remains  
453 open. Our experimental data do not support predictions drawn from the dual-isotope  
454 conceptual model (Scheidegger et al., 2000) (Table 4), assuming stomatal conductance as a  
455 proxy for WU and photosynthetic capacity as a proxy for biomass growth. Following Roden  
456 and Siegwolf (2012), this disagreement might be grounded in (i) the differences in ambient  
457 humidity and leaf temperature for different thinning treatments; the  $\delta^{18}\text{O}$  enrichment of soil  
458 water could be a source of uncertainty, as previously discussed; (ii) that the ratio of vapour

459 pressure deficit (D) to  $g_s$  was not previously studied for *P. halepensis*, making it difficult to  
460 ascertain how potential humidity variation would translate into changes in  $g_s$ ; (iii) both  
461 isotopes being measured in tree rings, which could show dampened signals when compared to  
462 leaf  $\delta^{18}\text{O}$ ; (iv) the ambient humidity/D ratios between treatments and years not remaining  
463 constant - this may well be the greatest limitation on using this model in plots with different  
464 treatments; and (v) the drought signal (common in Mediterranean summers) in isotope  
465 composition in leaves might not be well represented in tree ring material because of shut-off  
466 periods in secondary growth.

467 Other studies in this species also found discrepancies between  $\text{WUE}_i$  from leaf-scale  
468 gas exchange measurements and from the integrated tree ring-based values (Klein et al.,  
469 2013a). To the best of our knowledge, our study is the first study in semiarid forests to  
470 compare WUE measurements at stand level, with thinning treatments, and  $\text{WUE}_i$  in tree rings  
471 as a time-integrated proxy of WUE during the growing season. Consequently, more studies  
472 will be needed to clarify to what extent stable isotopes are suitable tools in forest  
473 management.

## 474 **Conclusions**

475 While some of the effects of thinning have been pointed out in other studies, this  
476 paper introduces a novel contribution relating water use efficiency (WUE), as measured by  
477 sap flow sensors, and intrinsic water use efficiency ( $\text{WUE}_i$ ) derived from stable isotope  
478 analyses in the same trees in a Mediterranean Aleppo pine plantation subjected to different  
479 thinning intensities.

480 Thinning induced differences in tree growth rates and changed the tree growth-  
481 climate relationships, showing higher dependence in the non-thinned plot on monthly  
482 precipitation in the current year whatever the season, whereas tree growth in the thinned plot

483 was more related to the expected ecophysiological pattern, i.e. precipitation in May. These  
484 results are more consistent as time has elapsed since the first thinning in 1998, creating a  
485 significant improvement in forest resilience to climate changes. The earlier high-intensity  
486 thinning treatment (H98) had a higher initial growth rate than the similar 2008 thinning  
487 treatment (H), showing that the delay in the decision to thin hampers initial growth rates.

488           Measurements of stable isotopes in tree rings have been said to provide powerful  
489 information about the physiological and environmental factors that control WU and WUE<sub>i</sub>.  
490 However, our study indicated slight or no changes in WUE<sub>i</sub> after thinning, and only dry years  
491 seemed to enhance WUE<sub>i</sub>, especially in the high-competing stands (control). According to  
492  $\delta^{18}\text{O}$  data and the Sheidegger et al. (2000) model, this enhancement is due to an improved  
493 photosynthetic rate. These results do not corroborate the experimental ones when the sapflow  
494 technique was employed, in which thinned plots had higher water use and also higher WUE  
495 than control plots due to a significant increase in tree growth after thinning, which seemed to  
496 be underestimated when using WUE<sub>i</sub> estimated from  $\delta^{13}\text{C}$ . This conclusion points to the need  
497 for more detailed studies along these lines, incorporating a broader sample of years and  
498 species, before general assumptions can be made about changes in WUE in Mediterranean  
499 forests, as derived from adaptive treatments.

## 500 **Acknowledgements**

501           This study is part of the research projects “CGL2011-28776-C02-02, HYDROSIL”,  
502 “CGL2014-58127-C3-2, SILWAMED,” funded by the Spanish Ministry of Science and  
503 Innovation and FEDER funds, and “Determination of hydrologic and forest recovery factors  
504 in Mediterranean forests and their social perception,” supported by the Ministry of  
505 Environment, Rural and Marine Affairs. The authors are grateful to the Valencia Regional  
506 Government (CMAAUV, Generalitat Valenciana) and the VAERSA staff for their support in

507 allowing the use of the La Hundo experimental forest and for their assistance in carrying out  
508 the fieldwork. We express our gratitude to Professor R. Montes for constructive criticism and  
509 suggestions on an earlier version of the paper. The first author thanks the Mundus 17  
510 Programme, coordinated by the University of Porto (Portugal).

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697

698 Table 1. Forest structure variables in each plot studied. DBH is average Diameter at Breast  
699 Height. Adapted from Molina and Del Campo (2012) and Del Campo et al. (2014).

Treatment	Cover (%)	Density (trees ha <sup>-1</sup> )	DBH (cm)	Mean height (m)	Basal Area (m <sup>2</sup> ha <sup>-1</sup> )
Control (C)	84	1489	17.8	11.5	40.1
Low intensity (L)	68	744	21.2	12.2	27.2
Medium intensity (M)	50	478	21.7	11.3	18.2
High intensity (H)	22	178	20.4	12.2	9.4
High intensity-1998 (H98)	41	155	25.2	12.6	13.6

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701



702 Table 2. Mean values of Tree Ring Width (TRW, cm), intrinsic Water Use Efficiency (WUE<sub>i</sub>,  
 703  $\mu\text{molCO}_2/\text{molH}_2\text{O}$ ) and oxygen isotopic signature ( $\delta^{18}\text{O}$ , ‰). Mean values followed by the  
 704 same letter did not differ significantly either at short-term or at mid-term ( $p < 0.05$ ).

Time span	Thinning	Treatment (thinning intensity)	TRW*	WUE <sub>i</sub>	$\delta^{18}\text{O}$
Mid-term (1995-2010)	Before ('95-'97)	Control	0.09 (a)	98.5 (a)	28.7 (a)
		High98	0.06 (a)	97.4 (a)	28.9 (a)
	After ('08-'10)	Control	<b>0.06 (b)</b>	<b>94.4 (a)</b>	28.2 (a)
		High98	<b>0.17 (a)</b>	<b>88.7 (b)</b>	29.0 (a)
Short-term (2004-2010)	Before ('04-'07)	Control	0.05 (a)	100.4 (a)	28.2 (a)
		High	0.07 (a)	101.8 (a)	28.3 (a)
		Medium	0.06 (a)	96.4 (a)	28.1 (a)
		Low	0.05 (a)	100.5 (a)	28.2 (a)
	After ('08-'10)	Control	<b>0.07 (c)</b>	94.4(a)	<b>28.2 (b)</b>
		High	<b>0.42 (a)</b>	97.6 (a)	<b>29.3(a)</b>
		Medium	<b>0.19 (b)</b>	94.1 (a)	<b>29.4 (a)</b>
		Low	<b>0.12 (bc)</b>	92.1 (a)	<b>28.5 (ab)</b>

705 \* Mean and differences in TRW refer only to those trees sampled for isotopes analyses.

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708 Table 3. Mean values of Tree Ring Width (TRW, cm), intrinsic Water Use Efficiency (WUEi,  
709  $\mu\text{molCO}_2/\text{molH}_2\text{O}$ ) and oxygen isotopic signature ( $\delta^{18}\text{O}$ , ‰). Mean values between  
710 treatments followed by the same letter did not differ significantly. \* and \*\* indicate  
711 significant differences at  $p < 0.05$  and  $p < 0.01$  respectively.

Time	Year type	Treatment	TRW	WUEi	$\delta^{18}\text{O}$
Before thinning	Dry	Control	0.06 (a)	101.4 (a)	28.5 (a)
		H98	0.06 (a)	99.3 (a)	29.3 (a)
	Wet	Control	0.12 (a)	94.8 (a)	28.7 (a)
		H98	0.07 (a)	93.8 (a)	28.9 (a)
After thinning	Dry	Control	0.04 (b)**	100.7 (a)**	28.4 (b)**
		H98	0.33 (a)**	96.7 (b)**	29.8 (a)**
	Wet	Control	0.06 (b)**	97.3 (a)	28.2 (b)*
		H98	0.37 (a)**	94.2 (a)	29.5 (a)*

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716 Table 4. Results of the dual-isotope model (Scheidegger et al. 2000) after applied to our  
 717 experimental data (2009-2010): relative variation in  $\delta^{13}\text{C}$ ,  $\delta^{18}\text{O}$ , tree water use (WU) and  
 718 biomass growth between the different thinning treatments and the control. Up or downward  
 719 arrows represent increasing or decreasing values,  $\approx$  indicates insignificant changes.

Treatment	This paper's data (2009-2010)				Scheidegger et al. (2000)		
	Relative variation to the Control (treatment value / control value)				Theoretical variation expected for stomatal conductance, g (proxy for WU) and photosynthetic capacity, Amax (proxy for biomass growth)		
	$\delta^{13}\text{C}$	$\delta^{18}\text{O}$	WU	Biomass growth	$\delta^{13}\text{C}$ , $\delta^{18}\text{O}$ scenario (model input)	Model output	
						g	Amax
Low	1.004	1.022	1.109	1.972	b, c	↓, ↓	$\approx$ , ↓
Medium	1.003	1.060	1.054	2.333	b, c	↓, ↓	$\approx$ , ↓
High	0.994	1.044	3.592	8.938	c, d	↓, $\approx$	↓, ↓
High 98	1.020	1.037	2.347	3.668	b	↓	$\approx$

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722 **Figure captions**

723 Fig. 1. Temporal dynamics of mean Tree Ring Width (TRW, cm; a: 1960-2010, b: 1995-  
724 2010) and the residual chronologies (TRW index, c: 1960-2010, d: 1995-2010) in each  
725 treatment. Arrows indicate the thinning dates.

726 Fig. 2. Intrinsic Water Use efficiency (WUE<sub>i</sub>,  $\mu\text{molCO}_2/\text{molH}_2\text{O}$ ) by single years. a: Control  
727 and High-intensity thinning in 1998, b: Control and High-intensity thinning in 2008, c:  
728 Control and Medium-intensity thinning and d: Control and Low-intensity thinning. Data  
729 shown are average values  $\pm$  SD. A star near the axis indicates significant difference at  $p<0.05$ ;  
730 a double star indicates significant difference at  $p<0.01$ ; and a dot indicates marginal  
731 difference ( $p<0.1$ ) in that year.

732 Fig. 3. Tree-ring Oxygen isotopic signature ( $\delta^{18}\text{O}$ , ‰). a: Control and High-intensity thinning  
733 in 1998, b: Control and High-intensity thinning in 2008, c: Control and Medium-intensity  
734 thinning and d: Control and Low-intensity thinning. Data shown are average values  $\pm$  SD. A  
735 star near the axis indicates significant difference at  $p<0.05$ ; a double star indicates significant  
736 difference at  $p<0.01$ ; and a dot indicates marginal difference ( $p<0.1$ ) in that year.

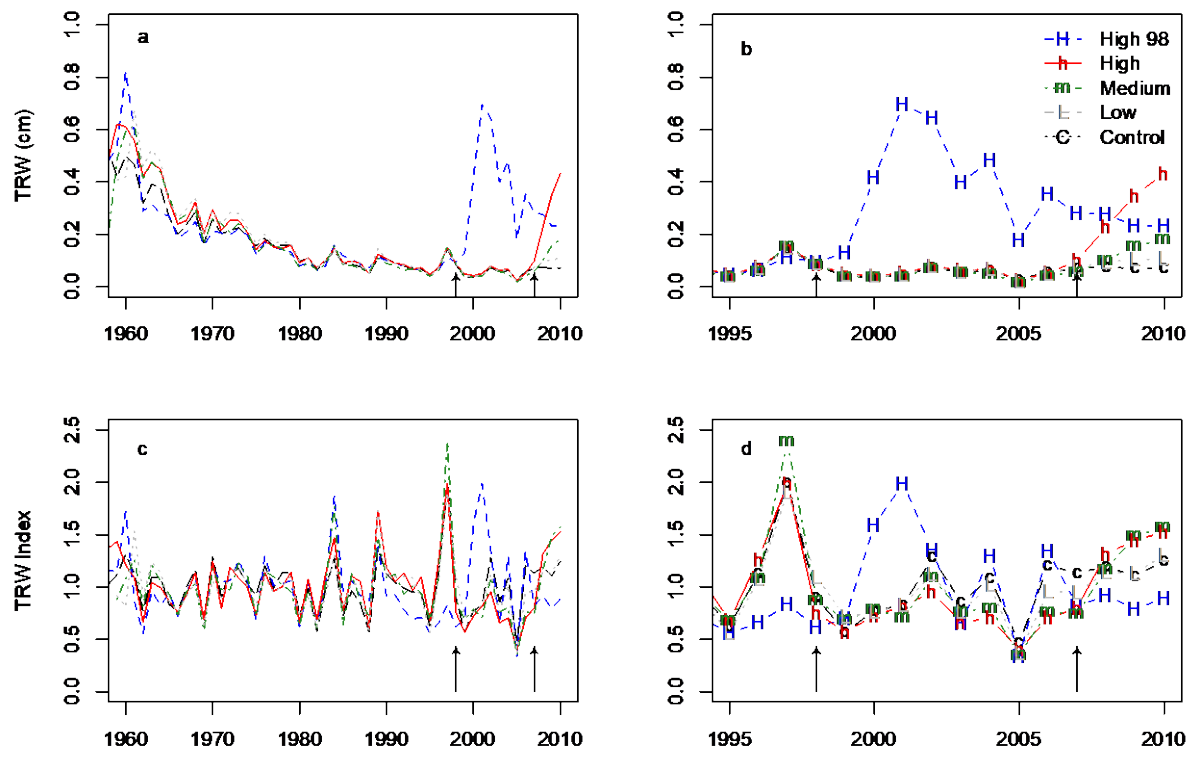
737 Fig. 4. Relationships between mean intrinsic water use efficiency (WUE<sub>i</sub>,  $\mu\text{molCO}_2/\text{molH}_2\text{O}$ )  
738 and total precipitation (Ppt, mm) for all treatments and years together. The black line and  
739 equation refer to all treatments together.

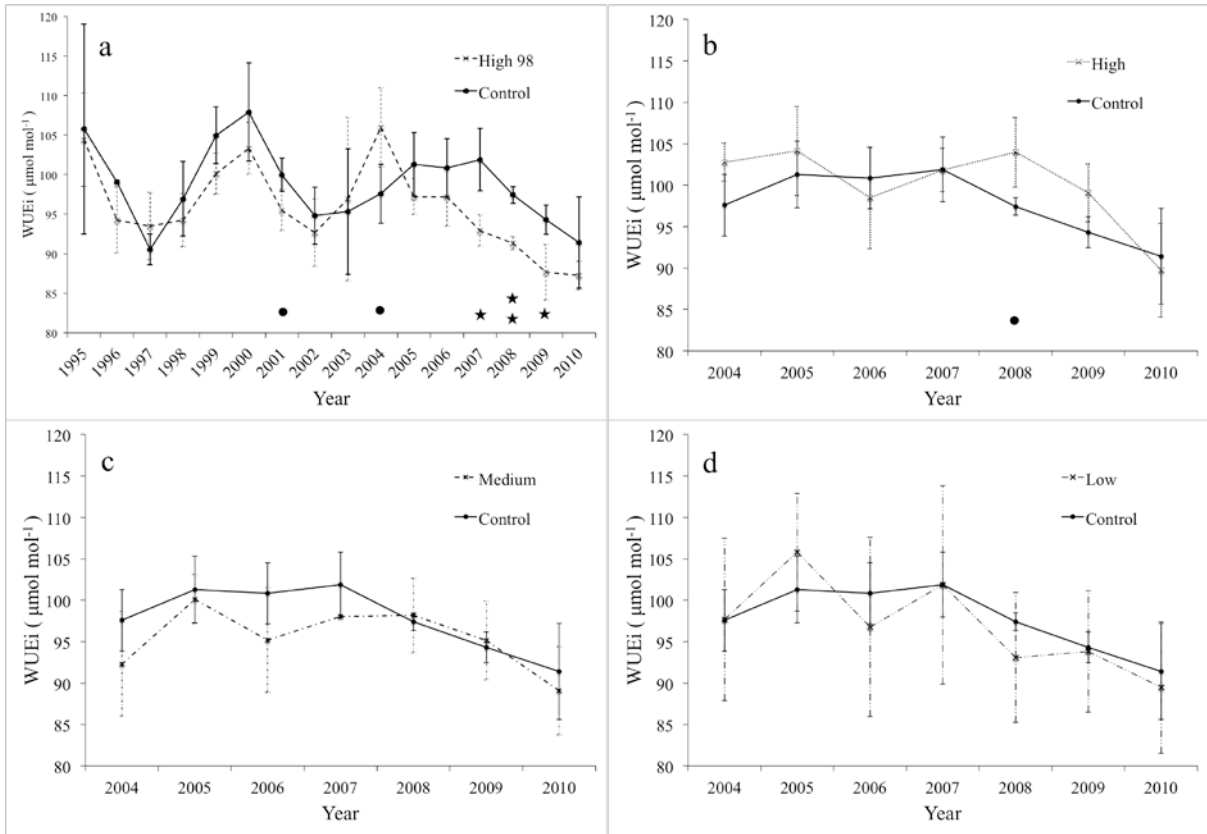
740 Fig. 5. Moving response functions of precipitation (ppt) from April of previous (prev) year to  
741 October of current (curr) year for a residual tree-ring chronology (TRWi). The window size is  
742 fourteen years; the windows have been offset by one year. a: Control plot; b: Thinned plot  
743 (H98). In each cell, the intensity of the colour represents the magnitude of the correlation

744 (positive or negative) between growth and monthly precipitation for that temporal window.  
745 Asterisks in a cell indicate significant correlations at  $p < 0.05$ . A clear change in trend can be  
746 observed in the thinned plot after treatment took place (1998). For a better interpretation of  
747 colours in this Figure, see the web version of this article.

748 Fig. 6. Mean annual WUE (g of dry weight increment / L of water transpired per tree) and  
749 water use (L of water transpired per tree) for the years 2009 and 2010 in the different  
750 treatments. Treatments with different letters indicate significant differences at  $p < 0.05$ .  
751 Different case letters mean different analyses.

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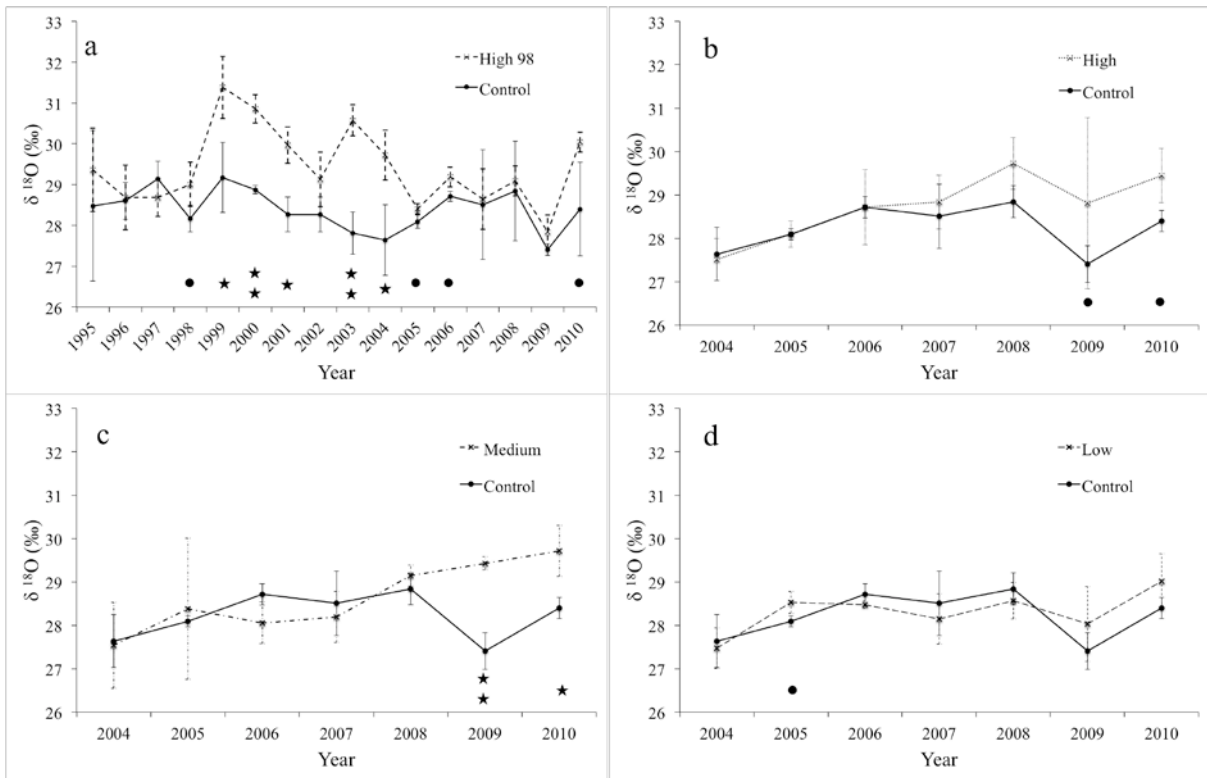
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758 Fig. 2.

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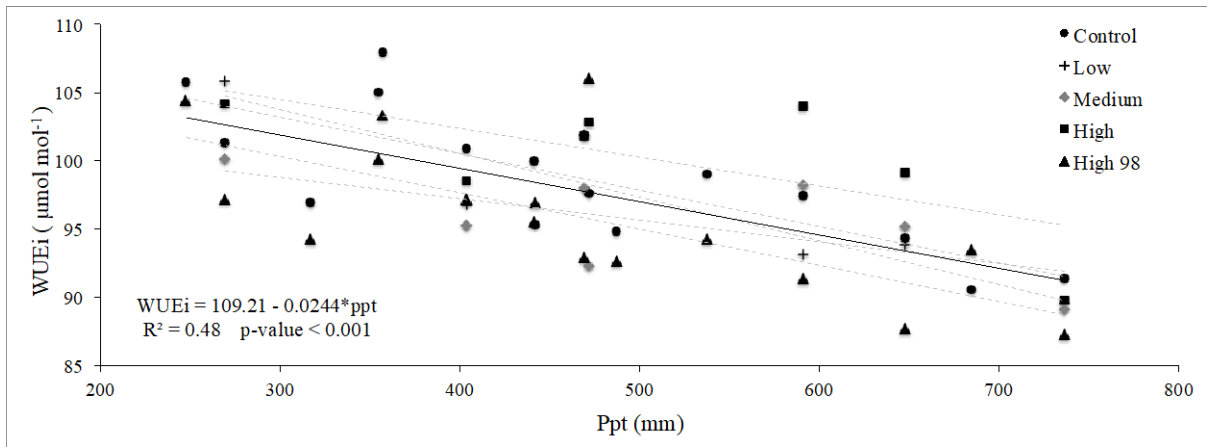
763 Fig. 3.

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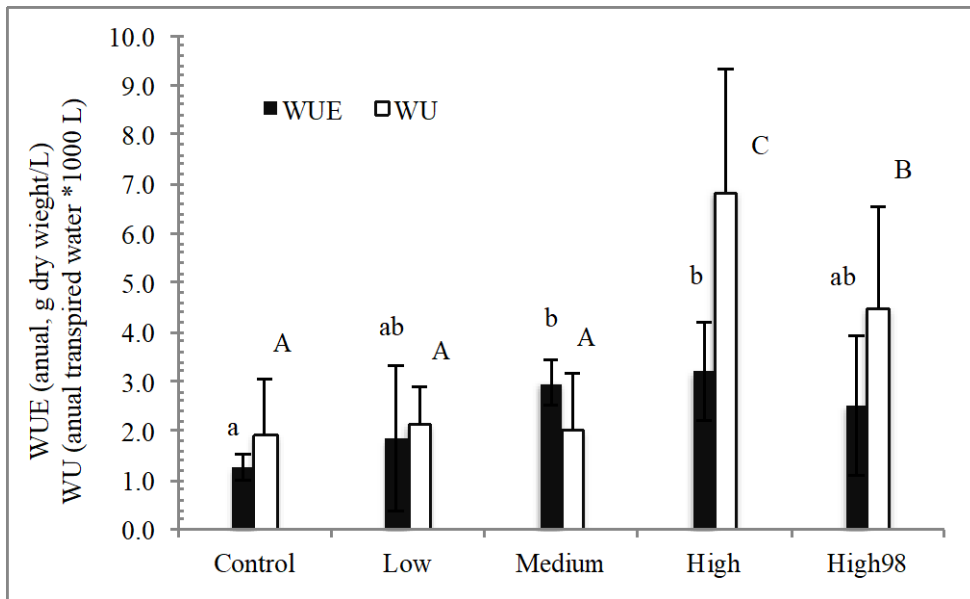
768 Fig. 4.

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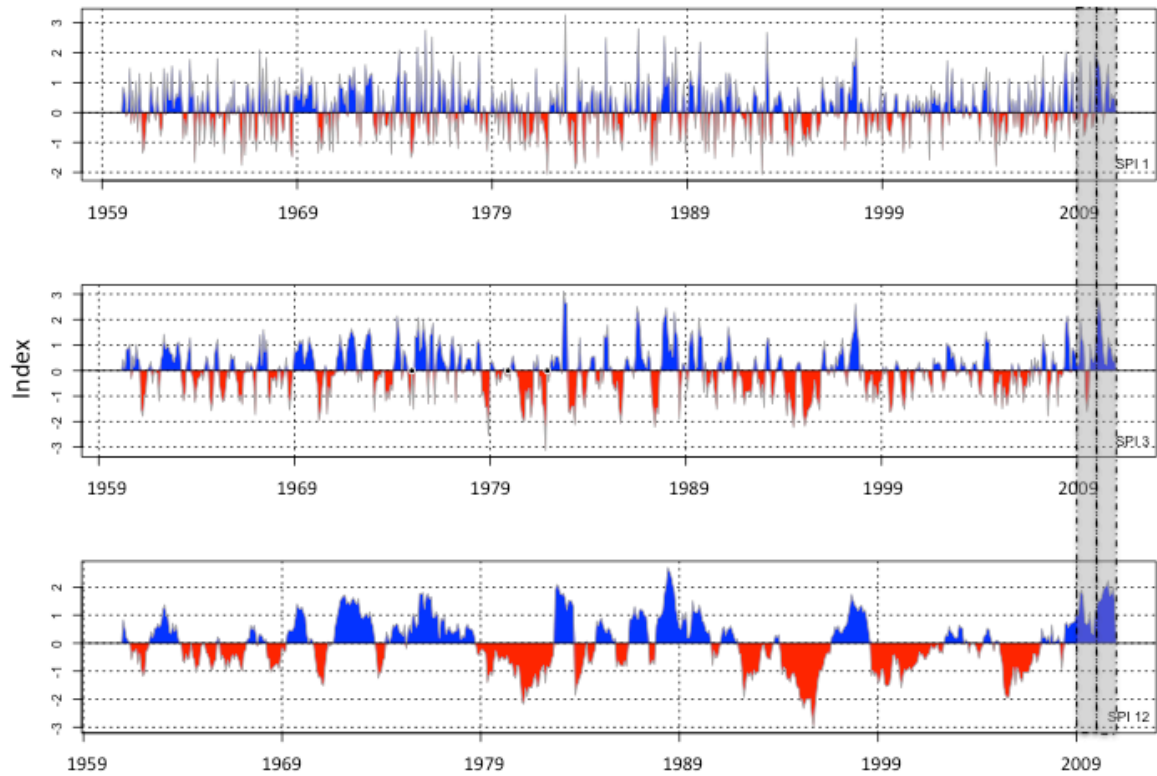
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776 Fig. 6.

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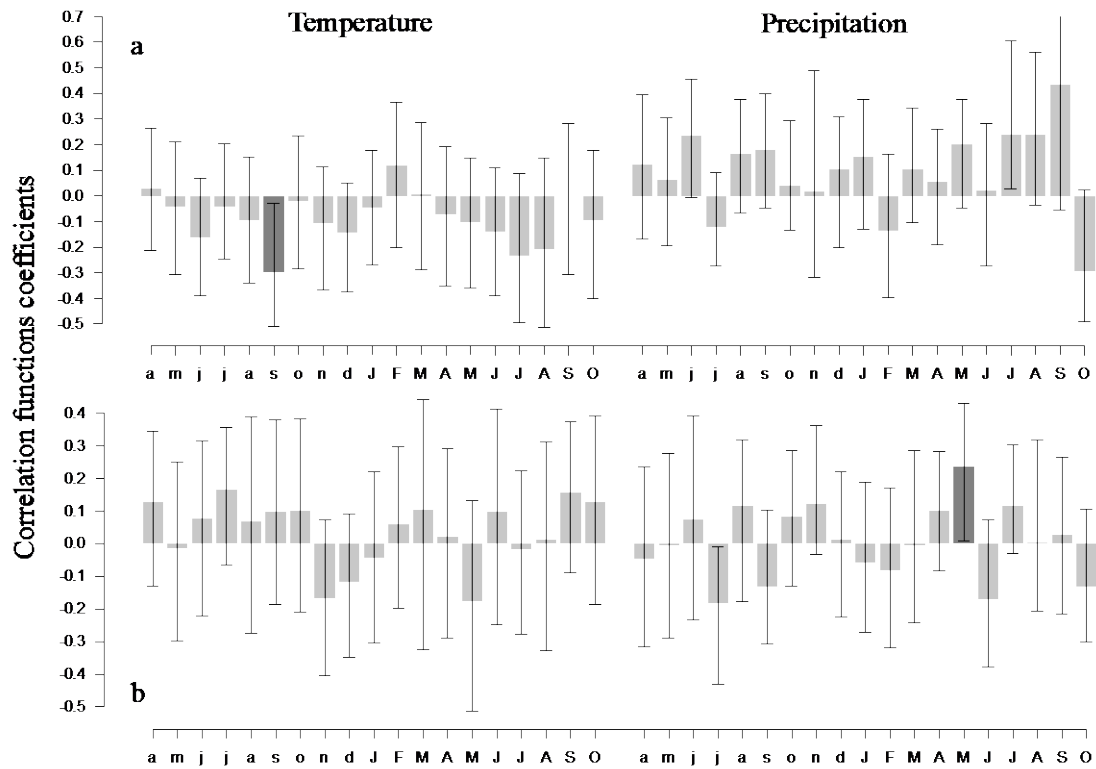
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779 **Supplementary materials**



780

781 Fig. 1S. Evolution of the drought index (SPI) at different time scales (1, 3, 12 months  
782 respectively) in the study area for the period 1961 to 2010. Grey-shaded area refers to years  
783 2009 and 2010 corresponding to the period of measurement of water use (tree transpiration) in  
784 this study.

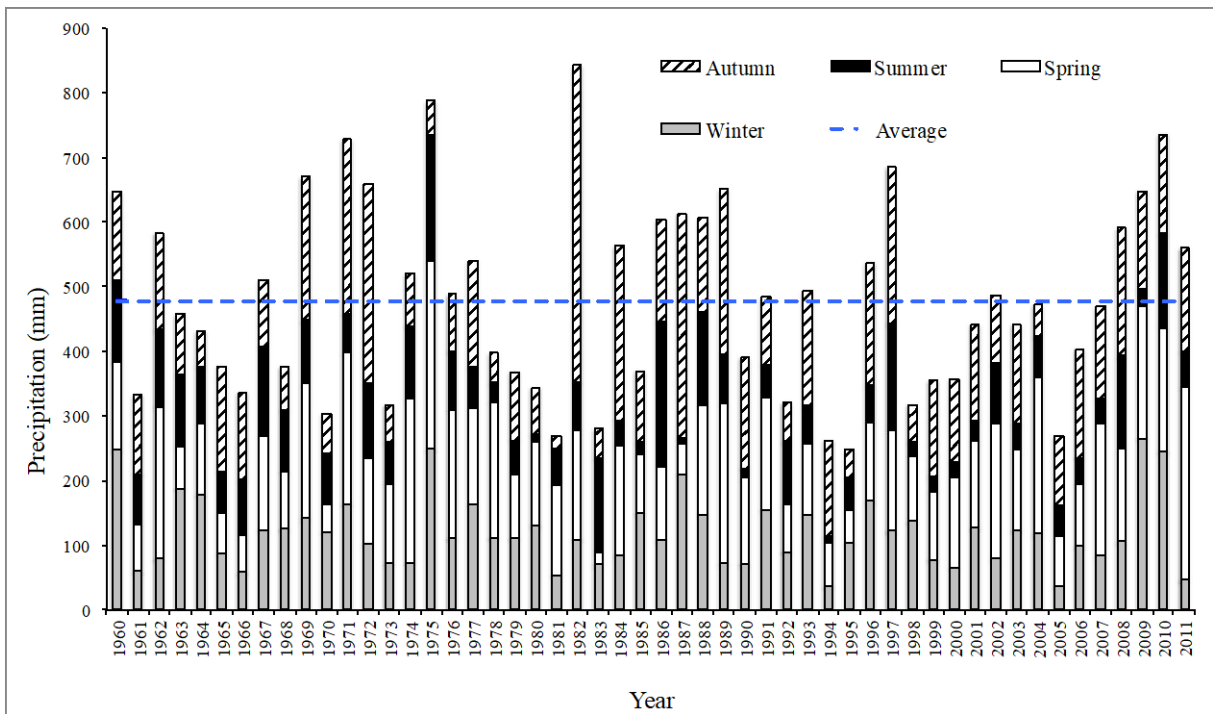


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787 Fig. 2S. Static bootstrapped correlation function (Zang and Biondi, 2013) for temperature and  
 788 precipitation from April of previous year to October of current year. a: Control plot, b:  
 789 Thinned plot (H98). The darker bars indicate a coefficient significant ( $p < 0.05$ ); the lines  
 790 represent the 95%-confidence interval. Lowercase letters represent the month of the previous  
 791 year (April to December) and uppercase letters, month of the current year (January to  
 792 October)

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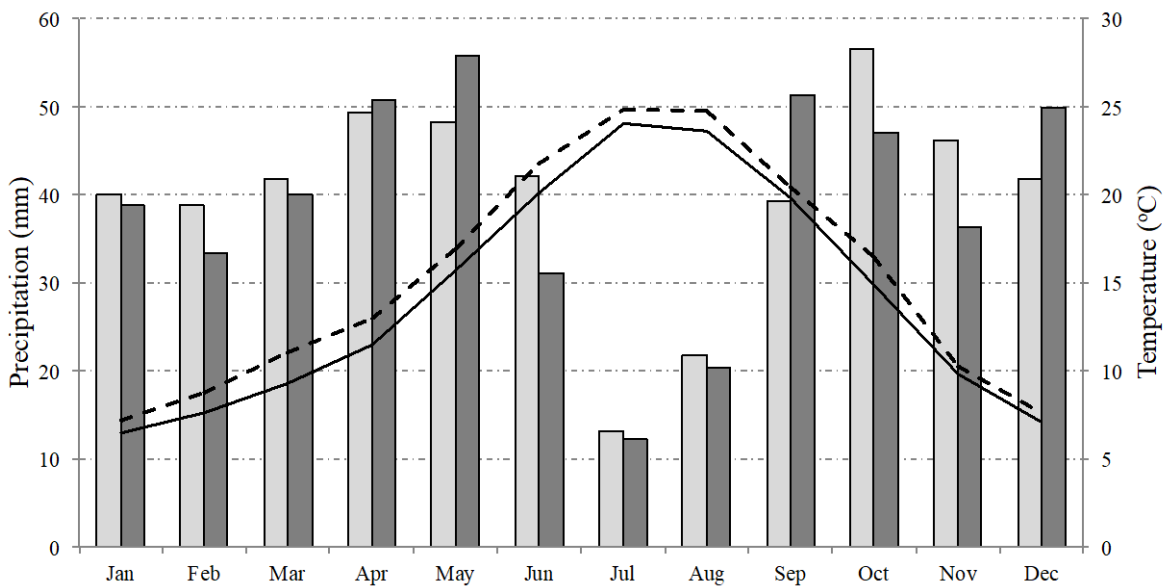


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795 Fig. 3S. Seasonal values of total precipitation during the period 1960-2011, following the  
 796 atmospheric circulation patterns as described in Pasho et al. (2011). Spring (April-May),  
 797 Summer (June-August), Autumn (September-November) and winter (December-March)

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801 Fig. 4S. Climatic diagram at La Hunde site. Grey and dark grey bars indicate total monthly  
 802 precipitation (mm) in the period 1960 to 2010 and 1995 to 2010 respectively. Continuous and  
 803 dashed line represent monthly mean temperature (°C) in the period 1960 to 2010 and  
 804 1995 to 2010 respectively.

805