

Mediterranean dwarf shrubs and coexisting trees present different radial-growth synchronies and responses to climate

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Abstract Due to their diversity and dominance in environmentally harsh sites, Mediterranean dwarf shrubs are a valuable tool to understand the consequences of climatic variability on radial growth in woody plants. We evaluate the dendrochronological potential of three Mediterranean dwarf shrubs versus three coexisting tree species inhabiting cold- (*Hormathophylla spinosa* vs. *Pinus sylvestris*), mesic- (*Ononis fruticosa* vs. *Abies alba*), and xeric sites (*Linum suffruticosum* vs. *Pinus halepensis*). Cross-sectional wood sections of the three shrub species and cores in the case of trees were visually cross-dated and ring-widths were measured and converted into residual growth indices. We used linear mixed-effects models to assess how growth indices respond to local factors and climatic variables. The radial growth of the three dwarf shrub species was more asynchronous, i.e., ring-width series differed among conspecific individuals, than that of coexisting tree species. Growth asynchrony was higher for *H. spinosa* than for *O. fruticosa* and *L. suffruticosum*. Similarly, the ring-width series of *O. fruticosa* and *L. suffruticosum* was strongly correlated with that of coexisting tree

species, while growth series of *H. spinosa* and *P. sylvestris* was not related at all. The growth of the three dwarf shrub species was influenced by the regional climatic conditions, but to a lesser degree than coexisting tree species. The highest responsiveness of growth to climate was observed in Mediterranean dwarf shrubs from xeric sites. However, local conditions are also major drivers of growth in Mediterranean dwarf shrubs as indicated by the stronger asynchrony in ring formation of these species as compared with coexisting trees, particularly in cold sites.

Keywords Annual ring · Chamaephyte · Dendrochronology · *Hormathophylla spinosa* · *Linum suffruticosum* · *Ononis fruticosa*

Introduction

Mediterranean dwarf shrubs or chamaephytes, i.e., plants that develop their renewal structures usually at a maximum height of 80 cm (Orshan 1982), constitute the dominant woody plant growth form in environmentally harsh (cold summits, dry steppes, etc.) or severely disturbed areas of the Mediterranean regions (Montserrat-Martí et al. 2011). For this reason, Mediterranean shrubs have been used as model systems to study seasonal variations in phenology, growth, and morphology (Palacio and Montserrat-

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Marti 2005), population dynamics (Eugenio et al. 2012), and plant–plant interactions (Soliveres et al. 2010). However, few dendrochronological studies have focused on Mediterranean dwarf shrubs by accurately cross-dating annually formed rings (but, see preliminary attempts in Camarero et al. 2010a; Genova et al. 2010; Soliveres et al. 2010; Eugenio et al. 2012). Since climatic models predict a temperature rise in mountainous regions and an aridity increase in drought-prone areas from the Circum-Mediterranean region (Christensen and Hewitson 2007), understanding how dwarf shrubs respond to such climatic oscillations might be of primary interest for the conservation of these communities.

Almost a century ago, Douglass (1920) recognized that trees form annual growth rings that can be cross-dated, i.e., synchronized among coexisting individuals of the same species subjected to similar climatic conditions, and thus changes in growth-ring features (e.g., width) will reflect climatic variability. Tree ring-width series has been used to reconstruct past climatic conditions in regions all around the world, but mainly in those regions showing a strong seasonality (Fritts and Swetnam 1989). More recently, it has been recognized that woody shrubs and dwarf shrubs also form distinct growth rings (Liphshitz and LevYadun 1986; Schweingruber and Poschold 2005; Bär et al. 2006; Schweingruber 2006), which can be visually cross-dated because of the dependence of secondary cambial growth on climatic oscillations (Bär et al. 2007, 2008; Li et al. 2008; Liang and Eckstein 2009; Forbes et al. 2010; Hallinger et al. 2010). However, while the cross-dating of tree rings is a well-established principle in dendrochronology, the analysis of annual growth rings in dwarf shrubs remains in its early days (Schweingruber et al. 2011). Problems associated with the adequate cross-dating of annual growth rings in dwarf shrubs reside in the heterogeneous wood development along the stem of these species (Kolishchuk 1990), the influence of the local conditions on annual wood formation (i.e., microsite dependence; Bär et al. 2006), and the short time span of shrub ring chronologies (Soliveres et al. 2010; Eugenio et al. 2012). These factors make it difficult to obtain precisely cross-dated and well-replicated ring-width chronologies in dwarf shrub species. However, studies performed in dwarf shrubs from Arctic and Alpine regions have demonstrated that those problems can be partially overridden and that shrub ring-width

chronologies reflect the influence of regional climatic conditions on growth (Rayback and Henry 2005; Bär et al. 2007, 2008; Liang and Eckstein 2009; Forbes et al. 2010; Hallinger et al. 2010).

In contrast to arctic and alpine regions, little is known about the dendrochronology and dendroecology of dwarf shrubs from the Mediterranean regions, although this growth form dominates regions where trees are scarce such as drought-prone steppes. In the Mediterranean region, dwarf shrubs tend to be short lived (Aragón et al. 2009; Eugenio et al. 2012) and they are strongly influenced by small-scale variations in the local environmental conditions (Soliveres et al. 2010). Although it has been demonstrated that secondary growth in Mediterranean dwarf shrubs responds to seasonal climatic conditions and thus their rings can be properly cross-dated (Camarero et al. 2010a), discovering if their ring-width chronologies reflect the influence of the local microenvironment more or respond more to regional climatic conditions still remains unexplored.

In the north-eastern Iberian Peninsula, where this study was performed, dwarf shrub species display diverse functional and phenological patterns of primary growth across pronounced climatic gradients (Palacio and Montserrat-Martí 2005, 2007). In a recent study, Gazol and Camarero (2012) showed that both cumulative primary (e.g., height) and secondary growth (e.g., basal diameter) of three Mediterranean dwarf shrub species inhabiting climatically contrasting sites were strongly influenced by the local microsite conditions. In this study, we aim to discover if the ring-width chronologies of these dwarf shrubs reflect the influence of inter-annual regional climatic conditions or are more driven by local factors. We do that by studying and carefully processing wood samples of these three dwarf shrub species dominating sites subjected to contrasting climatic conditions. Moreover, we compared the ring-width chronologies of the dwarf shrubs with those of coexisting trees by confronting the growth patterns and dendrochronological statistics of dwarf shrubs and trees. We hypothesize that: (1) local conditions influence more annual ring formation in dwarf shrubs than in trees; thus, we expect growth to be more synchronized among conspecific tree individuals than among conspecific shrub individuals; (2) the influence of the local conditions varies depending on the species and habitat considered; and (3) despite their relative microsite

dependence, ring-width chronologies of Mediterranean dwarf shrubs inhabiting stressful sites have dendrochronological potential since they reflect the influence of regional climatic conditions.

Materials and methods

Study area

The study was carried out in three different locations along a pronounced altitudinal (349–2,000 m a.s.l.) and climatic gradient situated between the Middle Ebro Basin and the Iberian System in Aragón, north-eastern Spain (Fig. 1). Specifically, we studied dwarf shrub and tree populations in the Javalambre cold site (high-elevation scrubland and grassland; 1,800–2,000 m a.s.l.), in the mesic site of Bernués (mild woodland; 900–1,000 m a.s.l.), and in the xeric site of Villamayor (semi-arid steppe; 300–400 m a.s.l.). Therefore, these three sites differ not only in their regional climatic characteristics, but also in their local topographic conditions (Fig. 1). To characterize the climatic conditions in each study site and to quantify the growth-climate relationships, we used data of meteorological stations located near the three sampling sites (Fig. 1). Specifically, we use data from the meteorological stations of Teruel (near Javalambre), Huesca (near Bernués), and Zaragoza (near Villamayor). We downloaded monthly mean temperature and total precipitation data for the period from 1970 to 2009 using the webpage of the Spanish Meteorological Agency (<http://www.aemet.es>). For each location, we calculated seasonal climatic variables by averaging (temperature) or summing (precipitation) for the previous autumn (September, October, November) and winter (December, January, February) before ring formation and for the current spring (March, April, May) and summer (June, July, August) of the year of growth.

Dwarf shrub and tree species

Three dwarf shrub species dominating different areas along this gradient were selected: *Hormathophylla spinosa* L. Küpfer (Brassicaceae) in the Javalambre cold site, *Ononis fruticosa* L. (Fabaceae) in the mesic site of Bernués, and *Linum suffruticosum* L. (Linaceae) in the xeric site of Villamayor. These species

were selected because (1) they are the dominant dwarf shrub species and form stable populations in each climatically contrasting site, (2) they belong to different families, and (3) they have recognizable annual growth rings (Fig. 2). A more complete description of the studied species can be found in Gazol and Camarero (2012). Three tree species dominating nearby sites (*Pinus sylvestris* L. in the cold site, *Abies alba* Mill. in the mesic site, and *Pinus halepensis* L. in the xeric site) were selected to compare their ring-width chronologies with those of the three dwarf shrubs. The field sampling and dendrochronological methods explained in the subsequent sections of this article refer exclusively to dwarf shrubs, but more details of sampling methods and processing of tree ring-width chronologies are available in Pasho et al. (2011).

Field sampling and dendrochronological methods

In each site, we randomly selected 20 mature individuals of each dwarf shrub species. All chosen individuals were removed and a transversal stem section was taken at 2 cm above the assumed root collar from the thickest stem, thus allowing exploration of the oldest stem part of each individual dwarf shrub. As previously done by Kolishchuk (1990), and to analyze intraplant growth variability and to detect locally absent rings along the main stem, we applied the “serial-sectioning” method (Bär et al. 2006). Accordingly, we selected ten individuals of each species with an age similar to the mode of each species (Table 1). In each of these individuals, five cross-sections of the thickest stem were taken at 5-mm intervals starting in the assumed root collar and going acropetally along the stem up to 20 mm above the root collar. Finally, the number of rings was counted in the five different cross-sections of each individual under a stereomicroscope at 10–50 magnification (M125 Leica Microsystems, Heerbrugg, Switzerland).

Transverse sections of stem samples were obtained in the field at the root collar and they were fixed in an ethanol-formaldehyde-acetic acid solution (ratio 90:5:5). Then, they were stained in the laboratory with a mixed solution of safranin (1 %) and methylene blue (1 %). After that, samples were carefully cleaned with three different ethanol solutions (50, 80, and 90 %) in three consecutive steps. Finally, three to four sections of each stem were mounted, permanently

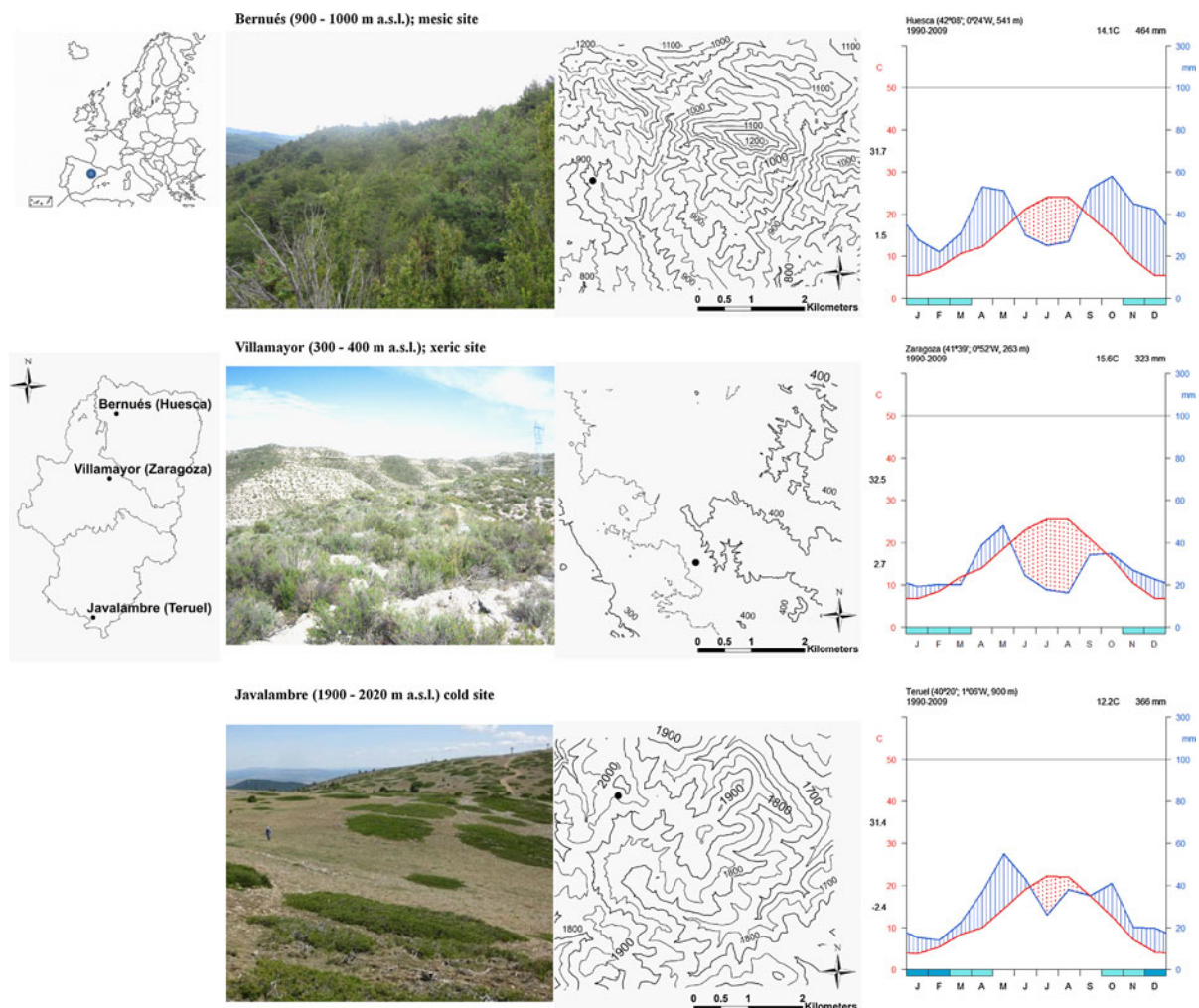


Fig. 1 Location of the study area and details of physiognomy, and the topographic and climatic conditions of each study site. We show the location of the study area (Aragón, north-eastern Spain) in Europe, as well as the location of each study site within the Aragón region. For each study site, the *photo* shows the characteristics of the landscape and dominant type of vegetation (Javalambre photo courtesy of A.I. García-Cervigón). The topographic map shows two types of contour lines: *solid ones* indicating changes in altitude of 100 m and *dashed ones* indicating changes in altitude of 50 m. The *solid black point* indicates the approximate location where dwarf shrubs

individuals were collected. The climatic diagrams were created with data of meteorological stations located near the three study sites using monthly data for the period from 1970 to 2009. The total precipitation (*blue line*, right axis) and mean temperature (*red line*; left axis) are shown. The area with vertical *blue lines* shows the months without water deficit, while the area with *red dots* shows the months with potential drought. The *light* and *dark blue squares* below the diagram show the months with potential and actual frosts. The mean annual temperature and the total annual precipitation are shown in the top right corner of the climatic diagram

fixed using the Eukitt[®] mounting medium, and examined under a light microscope (Olympus BH2; Olympus, Hamburg, Germany) within 10–30 min of staining at 100–500 magnification. Images were photographed with a digital camera (JVC 3.3 CCD GC-X3E; JVC, Yokohama, Japan) taking two pictures of each selected section. The quality of the mounted samples was based on the presence of the complete

section including the pith, the bark, and the entire xylem. The growth rings of the sections were examined, visually cross-dated, and measured up to the pith using the ImageJ software (available at <http://rsb.info.nih.gov/ij>; developed by W. Rasband, NIH). This was done for two radii per section. The visual cross-dating was checked using the program COFECHA (Holmes 1983), which calculates

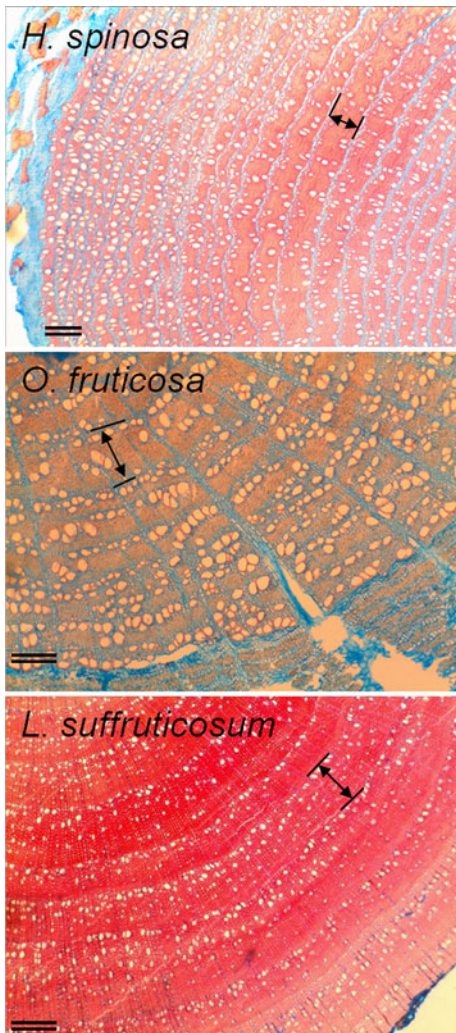


Fig. 2 Illustrative transversal sections showing the annual growth rings (delimited by *double-headed arrows*) and wood anatomic patterns of the three studied species. The *scale bars* are equivalent to 0.2 mm

correlations between ring-width series and a master chronology for a selected common period (1990–2009 for *L. suffruticosum* and *O. fruticosa*, and 1970–2009 in the case of *H. spinosa*). Finally, we selected 19 individuals of *H. spinosa*, 14 of *O. fruticosa*, and 16 of *L. suffruticosum* for further analyses (Table 2).

Dendrochronological analyses

To obtain a mean chronology of each individual and species, the cross-dated ring-width series was standardized and detrended using the program ARSTAN (Cook and Krusic 2005). First, we fitted a horizontal line (the mean value) through the ring-width series of each measured radius (Hallinger et al. 2010). Then, the ring-width data were divided by the fitted lines to obtain residual ring-width indices (RWI) that were subjected to autoregressive modeling and then averaged for each year using a biweight robust mean. We obtained mean residual chronologies for each individual and species of pre-whitened growth indices. The resulting mean chronologies of each species were described by calculating them for the common interval from 1990 to 2009 using three statistics widely employed by dendrochronologists: the first-order autocorrelation (AC1) of raw ring-width data (a measure of the year-to-year persistence in growth), the mean sensitivity (MSx) which is a measure of the relative year-to-year variability in width indices among consecutive rings, the mean between-individuals correlation (rbi) of the individual residual series, and the variance explained by the first principal component (PC1) (Fritts 1976). We also calculated the Expressed Population Signal (EPS) of the residual chronologies for the common period from 1990 to

Table 1 Descriptive growth and age statistics of the studied dwarf shrub and tree species

Site	Life form	Species	Ring-width (mm)	Age (years)
Cold site	Dwarf shrub	<i>Hormatophylla. spinosa</i>	0.08 ± 0.03 (0.01–0.27)	43 ± 9 (14–56)
	Tree	<i>Pinus sylvestris</i>	0.95 ± 0.04 (0.55–1.66)	138 ± 6 (86–219)
Mesic site	Dwarf shrub	<i>Ononis fruticosa</i>	0.18 ± 0.07 (0.02–0.49)	16 ± 3 (11–22)
	Tree	<i>Abies alba</i>	2.59 ± 0.11 (1.81–3.79)	73 ± 4 (50–121)
Xeric site	Dwarf shrub	<i>Linum suffruticosum</i>	0.15 ± 0.08 (0.01–0.53)	15 ± 3 (10–22)
	Tree	<i>Pinus halepensis</i>	1.29 ± 0.10 (0.52–2.47)	77 ± 7 (45–167)

The ring-widths and ages (mean ± SE) are displayed for each species. Minimum and maximum values for these variables are indicated between parentheses. In the case of dwarf shrub species, ring-widths were calculated for the common period from 1990 to 2009, while in the case of tree species, the period was variable (see Table 2; Pasho et al. 2011)

Table 2 Dendrochronological statistics of the studied species considering the common period from 1990 to 2009 (mean \pm SE)

Site	Species	No. individuals (no. radii)	Total time span	AC1	MSx	rbi	EPS	PC1 (%)
Cold site	<i>H. spinosa</i>	19 (33)	1970–2009	0.58	0.16	0.12	0.62	22.40
	<i>P. sylvestris</i>	14 (28)	1791–2006	0.82	0.18	0.40	0.91	44.23
Mesic site	<i>O. fruticosa</i>	14 (25)	1990–2009	0.27	0.25	0.24	0.69	32.44
	<i>A. alba</i>	20 (40)	1889–2006	0.76	0.17	0.38	0.89	45.40
Xeric site	<i>L. suffruticosum</i>	16 (26)	1990–2009	0.46	0.28	0.30	0.74	35.98
	<i>P. halepensis</i>	15 (29)	1843–2006	0.66	0.43	0.60	0.95	63.11
<i>F(P)</i> statistics comparing dwarf shrubs vs. trees				9.31 (0.04)	0.11 (0.76)	7.44 (0.05)	35.77 (0.004)	7.92 (0.05)

The first-order autocorrelation (AC1) was calculated for ring-width data, whereas the rest of variables (MSx mean sensitivity, rbi mean between-individuals correlation, EPS expressed population signal, PC1 variance explained by the first principal component) were obtained for residual ring-width chronologies. The last line indicates the statistics (*F* and *P* between parenthesis) of one-way ANOVAs comparing dendrochronological statistics between dwarf shrubs and trees

2009 since this statistic is commonly used to quantify the statistical quality of mean chronologies as compared with a perfect infinitely replicated chronology (Wigley et al. 1984).

In order to compare common patterns in secondary growth among coexisting dwarf shrubs and trees, we quantified the relationships between the residual chronologies of *H. spinosa*, *O. fruticosa*, and *L. suffruticosum* with those of coexisting tree species, namely *P. sylvestris*, *A. alba*, and *P. halepensis*, respectively, by means of the Pearson correlation coefficient. These comparative analyses were performed for the common period from 1990 to 2006.

Linear mixed-effects models of growth

We evaluated the relationship between growth and regional climate for each individual dwarf shrub and tree by means of linear mixed-effects models (LMM). We employed this approach to account for potential differences in growth among individuals (Ettinger et al. 2011) as compared with standard dendrochronological analyses that are based on mean chronologies of all individuals of the same species living in the same site (Fritts 1976). Therefore, we can study the influence of the regional climatic conditions on dwarf shrub and tree growth, while accounting for potential intra-specific growth asynchronies, i.e., differences between individuals, which can be interpreted as the influence of non-climatic local conditions on growth. We considered both individual dwarf shrub, or trees, and years as random effects to account for non-independence of data from the same individual or

within years (Crawley 2007). Seasonal climatic variables (see the aforementioned explanations) were used as fixed effects, i.e., as explanatory variables of growth indices (RWI). We used seasonal instead of monthly climatic variables because of the short length of the considered RWI chronologies (1990–2009) and to avoid the collinearity among climatic variables. We fitted several competing models containing different combinations of seasonal temperature and precipitation variables. Models were fitted using the maximum-likelihood estimation. We used Akaike's Information Criterion (AIC) to identify the "best-fitting" model for each dwarf shrub and tree species by comparing the null model (only intercept) with the different models fitted with different combinations of climatic variables ($AIC_{\text{null}} - AIC_{\text{best}}$). The linear model (1) showing the lowest AIC value and (2) containing the fewest parameters, when competing linear models showed differences in AIC scores lower than two, was selected following Burnham and Anderson (2002). Finally, we used linear regression analyses to study the relationship of each individual dwarf shrub and tree RWI with the climatic variables selected by the best-fitted LMM. We counted the number of individual dwarf shrubs and trees significantly influenced ($P < 0.1$) by the climatic variables. We used the *lme4* package in R for fitting LMMs (R Development Core Team 2011; Bates and Maechler 2010).

To disentangle if the dwarf shrubs and trees studied were more influenced by local (i.e., environmental heterogeneity) or regional factors (i.e., climatic variability), we considered the values of several dendrochronological statistics and the results of the LMMs.

Specifically, we considered that a higher dependence on local conditions will be indicated by a low synchrony in annual growth among individuals of the same species and a low sensitivity to climate variability, i.e., low values of the rbi, EPS, and MSx statistics as compared with other coexisting species. Similarly, the results of the LMMs will show a low influence of the regional climatic variables, and the proportion of individuals influenced by climatic conditions must be low. Finally, in order to compare the synchrony in secondary growth among coexisting dwarf shrubs and trees, we compared the dendrochronological statistics listed above of *H. spinosa*, *O. fruticosa*, and *L. suffruticosum* with those of coexisting tree species *P. sylvestris*, *A. alba*, and *P. halepensis*, respectively, by means of one-way ANOVAs.

Results

The three studied dwarf shrub species formed distinct growth rings and presented semi-ring porous wood type (Fig. 2). On average, *O. fruticosa* showed the widest annual rings (0.18 mm) and it was followed in decreasing order by *L. suffruticosum* (0.15 mm) and *H. spinosa* (0.08 mm) (Table 1). *H. spinosa* showed the longest life span, reaching in some individuals a maximum age of 56 years, followed by *O. fruticosa* and *L. suffruticosum* (the maximum age was 22 years in the two species) (Table 1). Among trees, *A. alba* followed by *P. halepensis* showed the widest rings (2.59 and 1.29 mm, respectively), whereas *P. sylvestris* was the most long-lived species (219 years). Our age estimates of dwarf shrub species were consistent since the serial sectioning performed at different distances from the root collar showed a constant number of annual rings along the stem for the three studied species (Fig. 3). These results indicate that transverse sections of shrub stem taken relatively close to the assumed root collar provide an accurate estimation of dwarf shrub age.

Overall, all dendrochronological statistics presented significantly lower means for dwarf shrubs than for trees other than MSx (Table 2). For instance, the mean rbi and PC1 values were 0.22 versus 0.46 and 30.3 versus 50.9 % for dwarf shrubs versus trees, respectively. For the dwarf shrubs, the lowest auto-correlation values in ring-width were obtained for

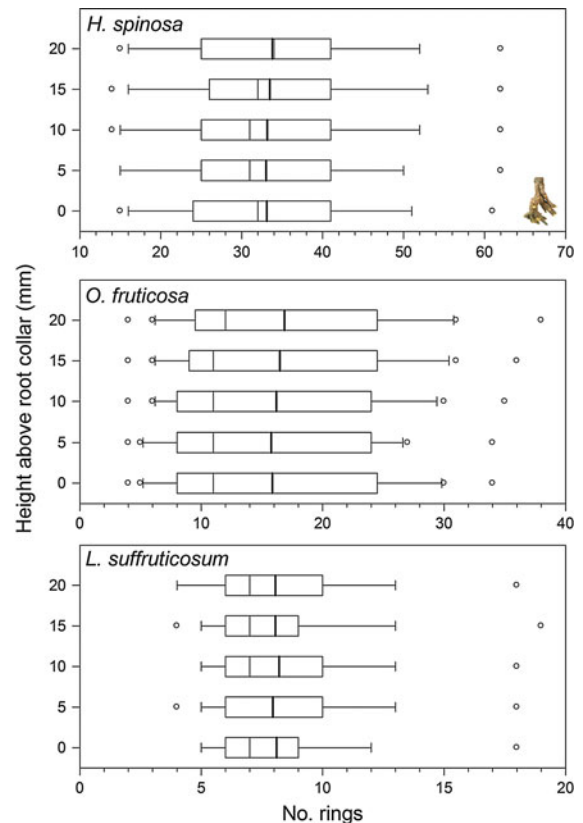


Fig. 3 Number of rings counted in sections taken at different heights above the root–shoot collar in the three dwarf shrubs studied: *H. spinosa*, *O. fruticosa*, and *L. suffruticosum*. The graph shows the mean number of tree-rings (box) as a function of the distance above the root collar (five different distances at increments of 5 mm) counted in ten individuals of each species. The lower and upper box boundaries correspond to the 25th and 75th percentiles, while the error bars located at the left and right sides of the box correspond to the 10th and 90th percentiles, respectively. The thin and thick lines within the boxes correspond to the median and the mean, respectively. In the upper graph, a view of a *H. spinosa* sample used to obtain ring estimates is displayed

O. fruticosa, whereas the highest ones were observed for *H. spinosa*. The highest MSx was obtained for *L. suffruticosum* followed by *O. fruticosa* and *H. spinosa*. The highest rbi, PC1, and EPS values were obtained for *L. suffruticosum* followed by *O. fruticosa* and *H. spinosa*.

The mean RWI of *H. spinosa* was positively, but non-significantly, correlated with that of *P. sylvestris* ($r = 0.29$, $P = 0.256$), whereas the RWI of *O. fruticosa* was strongly related to that of *A. alba* ($r = 0.58$, $P = 0.015$; Fig. 4). Finally, we found a highly significant and positive correlation in growth

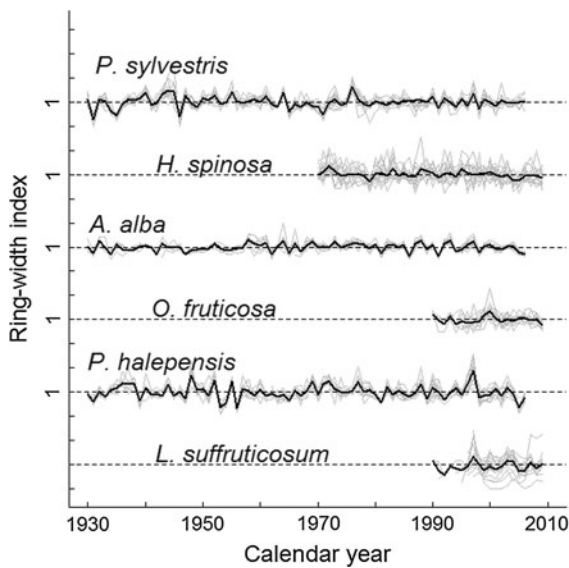


Fig. 4 Mean and individual ring-width residual chronologies of *Pinus sylvestris*, *Hormathophylla spinosa*, *Abies alba*, *Ononis fruticosa*, *Pinus halepensis*, and *Linum suffruticosum*. The black thick line shows the corresponding mean chronology of each species, while the gray thin lines show the individual series of each species

(RWI) between *L. suffruticosum* and *P. halepensis* ($r = 0.75$, $P = 0.001$).

In each site, the radial growth of the dwarf shrub and tree species was influenced by a different set of seasonal climatic conditions (Table 3; Fig. 5). The growth of

H. spinosa was positively correlated with previous winter temperature and negatively with current summer temperature, while the growth of *P. sylvestris* was positively related to current spring temperature and negatively to summer temperature. The growth of *O. fruticosa* was negatively influenced by the precipitation during the previous winter, but it was positively influenced by current spring precipitation. The growth of *A. alba* was also enhanced by current spring precipitation, but it was negatively related to summer temperature. Finally, the growth of *L. suffruticosum*, proved it to be the dwarf shrub species most sensitive to the climatic conditions (Table 3; Fig. 5). Specifically, the RWI of *L. suffruticosum* was enhanced by wet and warm conditions during the previous winter. A similar, but more complex, climatic model of RWI was obtained for *P. halepensis*.

Discussion

This study demonstrates the dendrochronological potential of Mediterranean dwarf shrubs since they form distinct annual growth rings. In spite of the fact that Mediterranean dwarf shrubs are short lived, it is possible to create reliable ring-width chronologies and relate them to regional climatic conditions. This hypothesis is also confirmed by the high correlation in secondary growth present between coexisting trees and dwarf shrubs, with the exception of the *H. spinosa*–*P. sylvestris* pair, which

Table 3 Sensitivity of the dwarf shrub and tree species to regional climatic conditions as assessed through the best-fitted linear mixed-effects models (LMM)

Site	Species	Best fit model	AIC _{null} – AIC _{best}	Proportion of individuals (%)
Cold site	<i>H. spinosa</i>	$+0.040 * T_{winter_{t-1}} - 0.045 * T_{summer_t}$	3.2	16
	<i>P. sylvestris</i>	$+0.080 * T_{spring_t} - 0.110 * T_{summer_t}$	4.9	64
Mesic site	<i>O. fruticosa</i>	$-0.001 * P_{winter_{t-1}} + 0.001 * P_{spring_t}$	3.4	27
	<i>A. alba</i>	$-0.070 * T_{summer_t} + 0.002 * P_{spring_t}$	7.3	64
Xeric site	<i>L. suffruticosum</i>	$+0.081 * T_{winter_{t-1}} + 0.004 * P_{winter_{t-1}}$	7.7	38
	<i>P. halepensis</i>	$-0.11 * T_{fall_{t-1}} + 0.17 * T_{winter_{t-1}} - 0.11 * T_{spring_t} - 0.12 * T_{summer_t} + 0.009 * P_{winter_{t-1}}$	24.4	93

The models were fitted to ring-width indices of individual dwarf shrubs or trees and considering as explanatory factors the seasonal climatic variables (T , mean temperature; P , total precipitation) for the previous year ($t - 1$) and the year of tree-ring formation (t). The best model was selected according to the Akaike's Information Criteria (AIC) by comparing the null model (AIC_{null}) with models fitted using climatic variables (AIC_{null} – AIC_{best}). Finally, each individual ring-width chronology of each species was regressed against the climatic variables selected by the LMM, and the proportion of individuals significantly influenced by the climatic variables is indicated

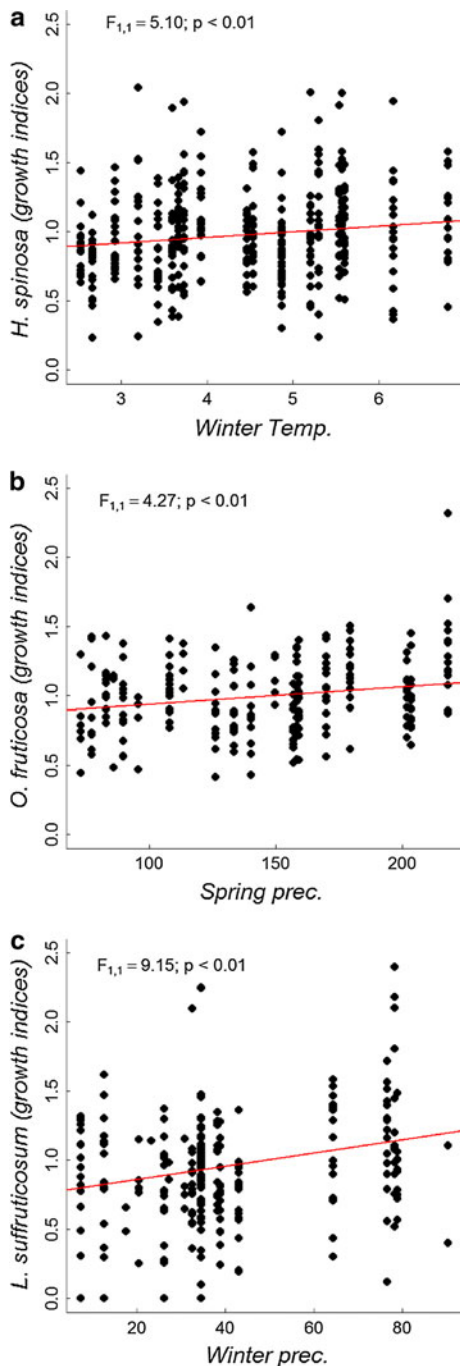


Fig. 5 Linear relationships between the individual residual ring-width indices (RWI) of *Hormathophylla spinosa* (a), *Ononis fruticosa* (b), and *Linum suffruticosum* (c) and previous winter temperature and current spring and previous winter precipitation, respectively. Additionally, the F value and associated probability levels measuring the relationship between the variables according to the linear mixed-effects models are shown

suggests that their growth patterns are influenced by regional climatic conditions. In agreement with our results, Liang and Eckstein (2009) showed that the radial-growth series of the alpine shrub *Rhododendron nivale* was correlated with that of a coexisting tree species (*Abies georgei* var. *smithii*). Finally, the results indicate that the dependence of the annual ring formation on the local environmental conditions, reflected here by the poor synchrony in growth among conspecific individuals, varies not only as a function of the species studied, but also probably due to the habitat conditions. Consequently, caution is needed when using Mediterranean dwarf shrubs as dendroclimatic models because they are short lived and their secondary growth is more influenced by the local conditions than that of coexisting tree species.

Mediterranean dwarf shrubs form annual growth rings as has been observed for different shrub species in semi-arid (Srur et al. 2011; Eugenio et al. 2012) and arctic and alpine regions (Bär et al. 2007; Xiao et al. 2007; Bär et al. 2008; Liang and Eckstein 2009; Forbes et al. 2010; Komac et al. 2011). All studied dwarf shrubs formed semi-ring porous wood, which agrees with previous anatomic studies on similar species (Schweingruber 2006; Schweingruber et al. 2011). Furthermore, the mean sensitivity and the autocorrelation values were comparable to those found for dwarf shrub species from cold regions (Xiao et al. 2007; Bär et al. 2008; Liang and Eckstein 2009). However, the mean correlation values with the master series and the EPS values, as well as the length of the ring-width chronologies, were lower for the three dwarf shrub species studied than in the aforementioned studies. In addition, no dwarf shrub chronology showed EPS values above the 0.85 threshold usually considered in dendrochronological studies (Wigley et al. 1984), while the chronologies of the three trees studied presented EPS values higher than 0.85. While the studies listed above have demonstrated the dendrochronological potential of arctic and alpine shrubs, the statistical evaluation of the synchronization in Mediterranean dwarf shrubs may be very difficult due to the short life span of some species.

Excluding *H. spinosa* in Javalambre, the chronology of which spanned almost 40 years (1970–2009), the studied Mediterranean dwarf shrubs tend to be short lived. The longevity of *H. spinosa* is in accordance with previous knowledge about the Brassicaceae family that indicates that shrubs and

dwarf shrubs of this family are slow-growing and long-lived (>50 years) species (Schweingruber 2006). In fact, this family includes Mediterranean woody species with dendrochronological potential living in semi-arid areas such as *Vella pseudocytisus* (Genova et al. 2010) and other species inhabiting mountainous areas subjected to short growing seasons (Schweingruber 2006). The short chronologies of *L. suffruticosum* and *O. fruticosa* are also in accordance with previous knowledge about Mediterranean dwarf shrubs from semi-arid areas, which indicates that they are short lived (Eugenio et al. 2012). This trait of dwarf shrubs living in semi-arid steppes might be linked to the huge amount of resources they allocate to flowering structures to increase their reproduction and recruitment success when climatic conditions are favorable (Aragón et al. 2009). Despite the low lifespan of these dwarf shrubs making it difficult to build long and reliable ring-width chronologies as indicated before, this study indicates that the secondary growth of Mediterranean dwarf shrubs and coexisting tree species responds to regional climatic conditions.

We found that the growth series of *L. suffruticosum* and *O. fruticosa* was well correlated with the growth series of the coexisting trees *P. halepensis* and *A. alba*, respectively. However, our results also showed that the growth series of *H. spinosa* and *P. sylvestris* was poorly correlated. Moreover, the ring-width chronologies of the three dwarf shrubs studied showed a much lower consistency in growth between individuals than nearby tree populations, indicating that secondary growth is more asynchronous in dwarf shrubs than in coexisting trees. This stresses the importance of preparing wood samples through adequate histologic methods (e.g., transversal sections in a sub-sample of individuals) and performing an accurate visual cross-dating of shrub wood samples, followed by a rigorous statistical checking of shrub ring-width series. For instance, Bär et al. (2008) demonstrated that an adequate cross-dating of annual ring series of dwarf shrubs allows controlling for microsite dependence and heterogeneous wood anatomy.

In addition, our findings suggest a high level of within-site (e.g., microenvironmental conditions) and plausibly intra-plant variability (e.g., plant architecture and resource allocation) in radial growth as has also been observed in arctic and alpine shrubs (Havström et al. 1993; Rayback and Henry 2005; Bär et al. 2007). The comparison of the three dwarf

shrub species indicates that synchrony in annual ring formation is lower for *H. spinosa* than for *O. fruticosa* and *L. suffruticosum*. The poorly synchronized growth of *H. spinosa* may indicate a strong influence of local conditions on its annual ring formation, and points to the microsite dependence of this species. Probably, *H. spinosa* is able to override the climatic stressors of the Mediterranean mountains (e.g., cold and long winters), their meristems being more coupled to microclimatic conditions near the soil and experiencing higher temperatures than tree species such as *P. sylvestris*. Therefore, the dwarf shrub species may be less influenced by the seasonal climatic oscillations than the tree ones. Ring formation may also start earlier in *H. spinosa* than in *P. sylvestris*, and that may be the reason why the growth of *H. spinosa* is positively influenced by winter temperature. However, these conclusions need to be interpreted with caution due to the scarce phenological and physiologic knowledge of the processes and mechanisms related to radial growth in dwarf shrub species (but, see Camarero et al. 2010a).

Previous studies have shown that cambial resumption of *L. suffruticosum* and *O. fruticosa* starts from late winter to early spring and the highest rates of wood formation occur in spring (from February to March) and early summer (from April to June), respectively (Camarero et al. 2010a). Moreover, these results agree with the data on primary and secondary growth phenology of Mediterranean dwarf shrubs (Montserrat-Martí et al. 2011). For instance, shoot growth rates of species inhabiting xeric sites such as *L. suffruticosum* reach maximum values in May, while dwarf shrubs inhabiting mild and cold sites expand their shoot growing season until late spring or even early summer (Palacio and Montserrat-Martí 2005, 2006). Therefore, wet conditions during late winter (January and February) before the ring formation starts will be of primary importance for growth in early spring of species from xeric sites such as *L. suffruticosum* and also *P. halepensis* (Camarero et al. 2010b), whereas warm conditions in mid to late spring will enhance growth in species such as *O. fruticosa* and *A. alba* inhabiting mesic sites (Camarero et al. 2011). The negative correlation between late winter precipitation and the growth of *O. fruticosa* may be explained by the negative effect of inter-specific competition on growth. In mesic sites, tree growth is strongly influenced by the competition with shrubs and

grasses (Peterson and Peterson 2001), thus higher water availability may enhance tree growth and increase grass establishment leading to reduced shrub growth in those areas. This relationship can be also spurious, since it is well known that it is difficult to separate the influence of climatic conditions on plant growth from other factors such as historic management (Gimeno et al. 2012), and the site where *O. fruticosa* was sampled has been regularly grazed (Gazol and Camarero, pers. obs.).

We have used dendrochronological techniques to analyze annual growth rings in Mediterranean dwarf shrubs, demonstrating that the species studied in climatically contrasting sites displays different growth patterns. Our results indicate that the radial-growth series of dwarf shrubs is influenced by the regional climatic conditions, indicating that these species can be used as integrative whole-plant models, and relating xylem variables (ring-width, wood anatomy) to other plant traits (e.g., above- and belowground primary growth). However, our results warn against the indiscriminate use of dendrochronology for performing retrospective analyses of radial growth in Mediterranean dwarf shrub species. Firstly, these species should be screened looking for cross-datable rings given that they present serious methodological difficulties (histologic analyses, laborious cross-dating). Secondly, the dwarf shrub species shows lower coherency in growth among conspecific individuals than trees. This low coherency may be caused by a higher influence of local conditions on radial growth in dwarf shrubs than in the case of trees. Contrastingly, in xeric sites, regional climatic conditions seem to drive the growth of dwarf shrubs and trees in similar ways. Therefore, we conclude that the dendrochronological potential of Mediterranean dwarf shrubs is higher in the most climatically stressed areas such as xeric and cold sites than in mesic sites.

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