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## Blue is the fashion in Mediterranean pines: New drought signals from tree-ring density in southern Europe

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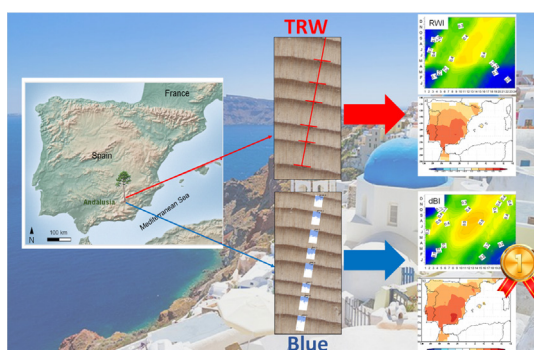
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### HIGHLIGHTS

- Blue intensity variables are sensitive indicators of drought events in low altitudes.
- BI variables showed more stability than raw ring-widths under extreme droughts.
- BI variables from Mediterranean conifers are reliable proxies for drought reconstructions.
- The use of BI and RW allows evaluating the impact of warming and water availability on pines.

### GRAPHICAL ABSTRACT



### ARTICLE INFO

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### ABSTRACT

Long-term records of tree-ring width (TRW), latewood maximum density (MXD) and blue intensity (BI) measurements on conifers have been largely used to develop high-resolution temperature reconstructions in cool temperate forests. However, the potential of latewood blue intensity (LWBI), less commonly used earlywood blue intensity (EWBI), and delta (difference between EWBI and LWBI, dBI) blue intensity in Mediterranean tree species is still unexplored. Here we developed BI chronologies in moist-elevation limits of the most southwestern European distribution of *Pinus nigra* subsp. *salzmannii* Arnold. We tested whether BI variables derived from tree rings of black pine are better proxies than ring-width variables to reconstruct long-term changes in climatic factors and water availability. For this we applied correlations and regression analyses with daily and monthly climate data, a spatial and temporal drought index (Standardized Precipitation-Evapotranspiration Index-SPEI) and Vapour Pressure Deficit (VPD), as well as atmospheric circulation patterns: North Atlantic Oscillation (NAO), Southern Oscillation Index (SOI) and Western Mediterranean Oscillation (WeMO). We found a positive relation between black pine growth (RW) and temperature during the winter preceding the growing season. Among all variables LWBI and dBI were found to be more sensitive than TRW to SPEI at low-elevation site, with EWBI series containing an opposite climatic signal. LWBI and dBI were significantly related to June and September precipitation at high-elevation site. Winter VPD was related with higher EWI and LWI series, whereas dBI and EWBI were related with January SOI and February NAO. We confirm the potential of long-term dBI series to reconstruct climate in drought-prone regions. This novel study in combination with other wood anatomical measurements has wide implications for further use of BI to understand and reconstruct environmental changes in Mediterranean conifer forests.

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## 1. Introduction

Knowledge on past changes in climate factors is limited by the available instrumental records. Some of these records reach back to mid- to late 1600, but for most areas systematic and continuous recording were not common until the mid-19th century (Overpeck et al., 2011). Therefore tree-ring records provide an invaluable year-to-year archive on paleoclimatic conditions and are - often in combination with other proxy data - used to reconstruct past climate (Carlón Allende et al., 2021; Williams et al., 2021) and effects of geological events, such as volcanic eruptions (Camarero and Ortega-Martínez, 2021; Tichavský et al., 2021). Moreover, tree-ring variables are indicators used in ecological studies, e.g., to study effects of extreme climate conditions, such as heatwaves and drought, on the physiological functioning of trees (Sánchez-Salguero et al., 2017; Sass-Klaassen et al., 2016). The fact that trees can live for centuries and sometimes millennia, makes them a perfect source to extract long-term information on changes in past climate and growing conditions in regions worldwide (Babst et al., 2018).

Year-to-year changes in maximum latewood density (MXD) in tree rings of conifer species has been found to be a better proxy than tree-ring width (RW) for reconstructing temperature (Yin et al., 2021), especially at higher latitudes with cool climate (Grudd, 2008; McCarroll et al., 2013). This is due to both a strong positive relation between MXD and the (summer) temperature-dependent growing season length, and to the lack of autocorrelation in the MXD series (Briffa et al., 2016; Rydval et al., 2015; Wilson et al., 2016). Up to now, only few studies have used MXD series to assess growth responses of conifer species under drought-limited conditions (López et al., 2021; Pompa-García et al., 2021). A recent study by Camarero and Hevia (2020) indicated the large potential to use tree-ring density to evaluate growth resilience to extreme drought events which is essential to evaluate future effects of global change on tree growth in drought-prone regions.

Although MXD records have proven to be a strong climate proxy, their wide application is hampered by the relatively time consuming and costly implementation of measuring wood density with X-ray scanning, or calculating the values based on wood anatomy using image analyses techniques (Björklund et al., 2019). Finding less expensive parameters that could provide robust data similar to MXD would facilitate dendroclimatic studies also for less equipped laboratories (Wilson et al., 2014). Blue intensity (BI) turned out to be a reliable and robust alternative. The BI is the intensity of reflectance of the blue channel light from a wood (core) image (McCarroll et al., 2002). Maximum reflectance of the blue light measured as “minimum blue intensity” from the latewood of conifer tree rings was found to be very similar to MXD values measured using X-ray densitometry (Björklund et al., 2014; Campbell et al., 2007). The BI values were initially linked to the main cell-wall components, cellulose, hemicellulose and lignin (Björklund et al., 2014; Wilson et al., 2014), but recent findings suggest that they depend on cell-wall dimensions, rather than on the cell-wall composition (Björklund et al., 2021). Moreover, another study on conifer species indicated a close link between BI values and cell-wall and cell-lumen ratio (Ho and Thomas, 2021), indicating the relevance of BI for assessing tree hydraulic characteristics. BI series are currently used for several purposes, such as reconstructions of past hydroclimate variability (Seftigen et al., 2020) and dating of historic timbers (Wilson et al., 2017). They have also shown a high potential for timber provenancing (Akhmetzyanov et al., 2020). However, the usefulness of using BI series in ecological studies, and specifically in water-limited areas, needs to be explored (Arbellay et al., 2018).

The growth of trees under continental Mediterranean climate is limited by dry conditions during summer, and low temperature in winter (Camarero et al., 2010; Sánchez-Salguero et al., 2015). With the projected climate change and increase in frequency and intensity of droughts across the Mediterranean basin, many tree species are expected to suffer from growth decline, an increase in defoliation rates and ultimately a higher mortality (Sánchez-Salguero et al., 2017). The growth response of Mediterranean trees to climate-caused changes in environmental conditions has

been widely studied using dendrochronological techniques (Domínguez-Delmás et al., 2013; Fyllas et al., 2017; Natalini et al., 2015; Touchan et al., 2016). BI can potentially accomplish this information by adding specific insights on the timing of effects of climate factors and/or the consequences of climate extremes on tree hydraulics. For example, in mountain areas, where several growth-limiting factors are at play, and thus the signal in tree-ring widths is mixed, BI series could pinpoint the most relevant factor influencing tree hydraulic properties under drought-induced stress. The Baetic Mountain range in southern Spain is one of such areas, where climate factors complexly influence the growth of black pine (*Pinus nigra*) resulting in unstable climate-growth relationships (Domínguez-Delmás et al., 2013; Sangüesa-Barreda et al., 2019). Black pine is the dominant species covering large areas of especially in southeast of this mountain range, and reaching high elevations close to 2000 m a.s.l. (Creus, 1998).

In this study, we used chronologies of several BI variables derived from tree rings of black pine from the drought-prone Mediterranean Baetic Mountains to test their potential to assess complex climate-growth relationships and serve - together with ring widths - as proxies to reconstruct past changes in climate factors, and to evaluate the effects of predicted increasing drought frequency. The long-span ring-width series (ring width (RW), earlywood width (EW), latewood width (LW)) and blue intensity (earlywood blue intensity (EWBI), latewood blue intensity (LWBI) and delta blue intensity ( $dBI = LWBI - EWBI$ )) of black pines in southeastern Spain, were tested for their sensitivity to local and regional daily and monthly climate. This was done by calculating climate - growth relationships between all ring-width and BI derived data and corresponding climate factor, including drought index and atmospheric circulation patterns. We aim: i) to use a combination of BI and RW related variables to quantify complex climate-growth relationships under Mediterranean climate making use local daily and monthly climate data, ii) to quantify resilience parameters of growth and wood density to climate extremes, and iii) to test the potential of BI-based climate reconstruction for drought-prone regions, in this case a well-studied area for developing long-term dendroclimatic reconstructions (Dorado Liñán et al., 2015; Esper et al., 2020).

## 2. Material and methods

### 2.1. Study site and regional climate data

The study area is located in Cazorla mountains, which are part of the Cazorla, Segura de Las Villas Natural Park located in the east of Andalusia, southeast of Spain and belong to the Baetic Mountain range (Fig. 1a). It is a large, protected area covering in total 2099.21 km<sup>2</sup> under Mediterranean climate with summer droughts and a significant difference between daily maximum and minimum temperatures (Tíscar, 2004). Most of the 1100 mm of annual precipitation (Table 1) is recorded in April, November and December, while July and August are the driest months (Fig. 1a). Average annual temperature is 11.7 °C, with a minimum of 4 °C in January and a maximum of 21 °C in August. For the last 100 years there was no trend in the annual amount of precipitation, but a significant increase in mean annual temperature (Fig. 1b, upper panel) and in the frequency and intensity of drought events (Fig. 1b, lower panel) have been observed. The bedrock is mainly formed by limestone and dolomites.

To assess the climate effect on tree growth and density, gridded daily E-OBS temperatures, and precipitation data with 0.1° grid was downloaded (Cornes et al., 2018). This data was further used for calculation of daily SPEI (Standardized Precipitation Evapotranspiration Index) values following Vicente-Serrano et al. (2010) and using the SPEI package (Begueria and Vicente-Serrano, 2017).

To test the potential of using BI for spatio-temporal analyses, and to understand the effects of vapour pressure deficit (VPD) and atmospheric circulation patterns on radial tree growth, given their wide spatial scale of influence (Stenseth et al., 2003), we considered the North Atlantic Oscillation (NAO), the Western Mediterranean Oscillation index (WeMO), and the Southern Oscillation Index (SOI). The NAO is a north-south dipole that determines the position of the Icelandic low pressure and the Azores

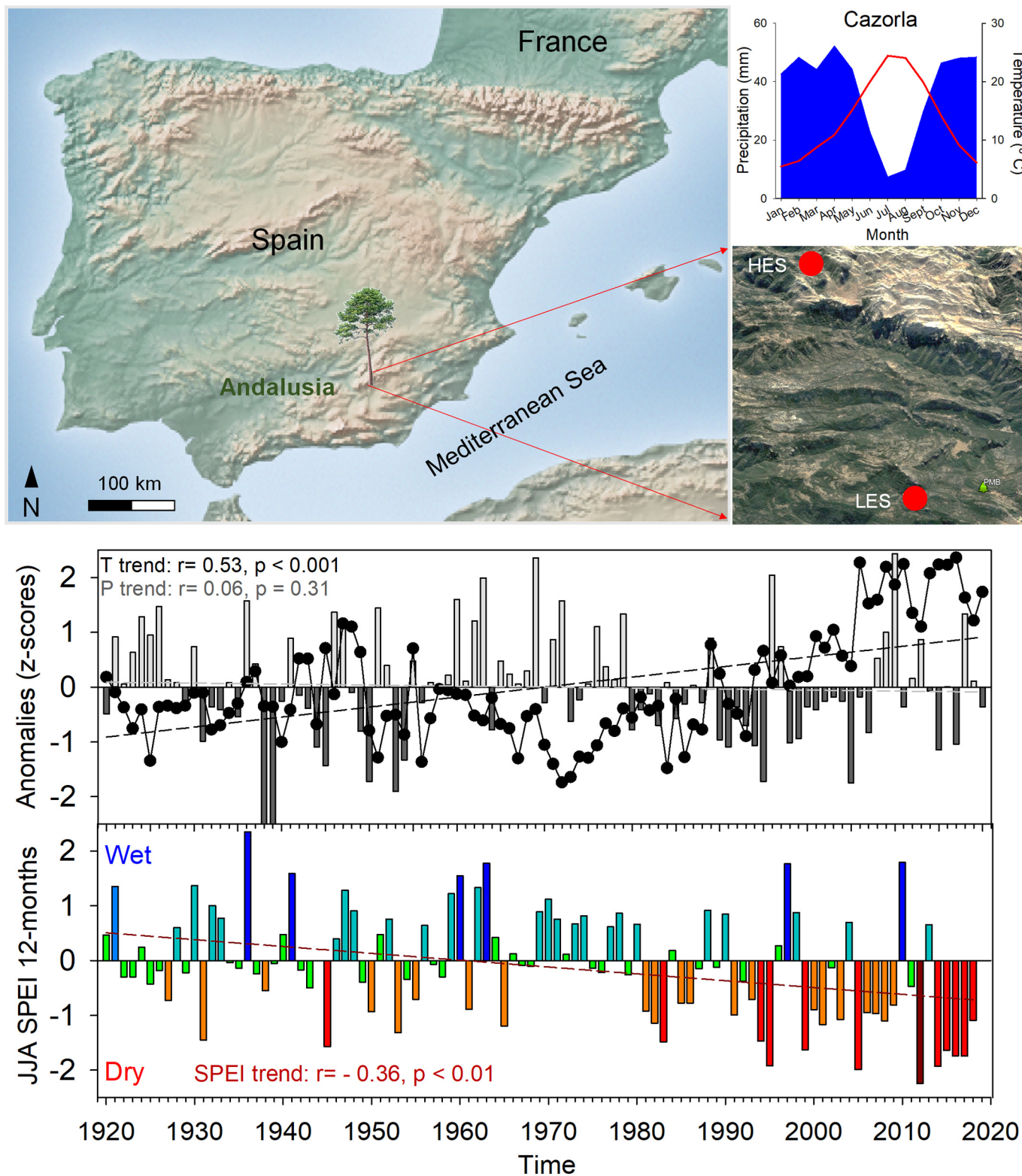


Fig. 1. Study site and climate data. a) Map showing the geographic location of the studied plots (red dots) together with the climate diagram representative for the study area; b) 100-year records of changes in annual precipitation (P) and mean temperature (T) expressed as the anomalies (standardized values) of total annual precipitation (P), mean annual temperature (T). The lower panel illustrates the temporal Standardized Precipitation Evapotranspiration Index (June–July–August SPEI 12-months scale); the SPEI is classified in three categories: orange/red (dry conditions); light/dark (wet conditions); green (moderate conditions), respectively.

high-pressure systems, and therefore the climatic patterns in southwestern Europe (Steele et al., 2001). The NAO has a strong influence on winter climate in the Iberian Peninsula (Camarero et al., 2022; Hurrell et al., 2003). The WeMO is the synoptic framework of the western Mediterranean basin

and its vicinities (Martin-Vide and Lopez-Bustins, 2006). The SOI reflects the effects of the El Niño Southern Oscillation (ENSO), and it is a standardized index of large-scale pressure differences between the western and eastern tropical Pacific and is known to be correlated with variations in global

**Table 1**

Features of the two study sites located at southern Europe in Sierra de Cazorla, Segura y Las Villas Natural Park (Low-elevation site: Linarejos and High elevation site: Navanoguera).

Variable	Low-elevation (LES)	High-elevation (HES)
Latitude N	37° 55'	37° 55'
Longitude	2° 54' W	2° 48' W
Elevation (m a.s.l.)	1191	1585
Mean diameter at breast height (cm)	49	59
Mean annual temperature (°C)	11.7	11.7
Maximum temperature (°C)	21	21
Minimum temperature (°C)	7	7
Annual precipitation (mm)	1100	1100
Bedrock type	Limestone	Limestone

precipitation patterns (Ropelewski et al., 1996). Several climate-growth studies in northern Spain have shown a major influence of NAO on spring conditions (Camarero, 2011; Rodó et al., 1997), and of SOI on the summer rainfall regime (Rozas et al., 2015). The WeMO has been previously linked to the differences in precipitation circulation of the western Mediterranean regions (Martín-Vide, 2002). The VPD, NAO and SOI monthly data were downloaded from the Climate Research Unit webpage (<https://crudata.uea.ac.uk/cru/data/pci.htm>); (Mitchell and Jones, 2005) and the WeMO from Climatology Group website at the University of Barcelona (<http://www.ub.edu/gc/wemo/>). The NAO, SOI and WeMO indices were calculated as a normalized pressure differences between Gibraltar (UK, in the south of the Iberian Peninsula) and Reykjavik (Iceland), between Tahiti (France) and Darwin (Australia), and between San Fernando (Spain) and Padova (Italy), respectively. NAO, SOI and WeMO indices show a high temporal variability with no significant correlations between them (Martín-Vide and Lopez-Bustins, 2006).

## 2.2. Dendrochronological methods

From 2014 to 2016, we sampled two black pine stands located at contrasting elevations at the Cazorla forest (see Table 1), one located at low elevation – Linarejos (1191 m a.s.l., hereafter LES) and a high elevation site, Navanoguera (1585 m a.s.l., hereafter HES). The largest and apparently oldest pine trees with no visible damage to crowns or stems were selected for sampling, and two to four 5-mm tree-ring cores perpendicular to the maximum slope were taken from each tree at breast height. In total 64 trees were sampled at LES and 63 trees at HES. Collected samples were air dried, mounted on wooden holders and cut using a sliding core microtome (Gärtner and Nievergelt, 2010). RW, EW and LW values were measured to the nearest 0.001 mm using a Velmex table (Velmex Inc., Bloomfield NY, USA) coupled to stereomicroscope (Olympus SZ60). Crossdating was done in two steps: visually and then statistically with PAST5 v. 5.0.615 (Knibbe, 2004) and followed standard dendrochronological procedures (Pilcher, 1990).

## 2.3. Wood density-blue intensity analysis

For the BI measurements, a subset of the entire core collection was used, i.e., cores without big cracks or discolorations. This subset included 14 trees from LES, and 11 trees from HES. Before BI measurements the core samples underwent chemical treatment to remove extractives (resin), which could alter the measurements and induce biases in the resulting data (Buckley et al., 2018). Next, they were immersed in acetone, which was renewed every 24 h to prevent saturation, for a total of 72 h, as this time was found to be sufficient to remove all the extractives (Rydval et al., 2014). Then, the samples were scanned with an Epson Expression 10000XL flatbed scanner at a resolution of 2400 dpi. Before scanning, the machine was calibrated using IT8 Calibration Target IT8.7/2 of SilverFast Auto 8, to ensure comparability of the results with other laboratories (Björklund et al., 2019). Moreover, the scanner was isolated from the ambient light by covering it with a box, fitted to the size of the scanner.

The measurement of BI values (EWBI and LWBI) was done using CooRecorder 9.0.1 (Larsson, 2014). To take into account colour differences between heartwood and sapwood of the black pine samples, dBI values were calculated as difference between EWBI and LWBI (Björklund et al., 2014). Since the raw BI data is inversely related to wood density (Campbell et al., 2007), obtained values were inverted by multiplying each value by  $-1$ , and then adding the constant 2.56 (related to light intensity scale 0–255) to remove any negative values. The constant was added in order to prevent negative blue intensity values, the specific value was chosen because the colour values are recorded with the maximum intensity set at 2.56 (Rydval et al., 2014).

## 2.4. Statistical analysis

Both ring-width related, and BI series were first averaged per tree and then detrended using a smoothed cubic spline (Cook et al., 1990) with a frequency response of 50 % at a wavelength of 32 years to remove low-frequency variations and emphasize high-frequency (year-to-year) ones. Master site chronologies for each variable were calculated by averaging (bi-weight robust mean) the obtained indices using 'dplR' package (Bunn, 2010) in R (R Core Team, 2021). The obtained standardized (index) chronologies of tree-ring variables are indicated as RWI, EWI and LWI.

The strength of the population signal in the chronologies of RWI, EWI and LWI was described by calculating dendrochronological quality statistics for the common period 1900–2014: the mean correlation between individuals ( $\bar{r}$ ), the first-order autocorrelation (AC) and the mean sensitivity (MS) of indexed series (Wigley et al., 1984). Climate-growth relationships between site chronologies of all measured ring-width and BI-related variables and selected local climatic factors and regional atmospheric circulation patterns for the period of 1951–2014 were calculated by means of Pearson's bootstrapped correlation analyses using *treeclim* package (Zang and Biondi, 2015) for monthly variables, and *dendroTools* (Jevšenak and Levanič, 2018) for calculations with daily data. Correlations between tree ring variables and monthly variables were calculated using a 14-months window, from September of the previous growing season to October of the current growing season (see Sangüesa-Barreda et al., 2019). The calculations with daily climate data were done to enhance the understanding of short-term effects of climate variables on tree growth (Jevšenak and Levanič, 2018) using a total window of 720 days from the previous to the current year of growth.

To assess differences in response to extreme events (droughts) between the measured ring variables, resilience components of the raw data (resistance – capacity of trees to buffer the drought event, resilience – growth after drought relative to that measured before the event, and recovery – growth in years after the drought event against the growth in the drought year) were calculated according to Lloret et al. (2011), considering a time window of 5 years prior to and after the drought event using *pointRes* package (van der Maaten-Theunissen et al., 2021).

In addition, to examine the difference in spatial extent and strength of the climatic signal between all measured tree-ring variables, field correlations analyses was performed following the methods proposed by Trouet and Van Oldenborgh (2013) using climatic factor which has shown the largest difference in response between the measured variables.

Correlation analyses identified those monthly climatic factors most strongly and significantly ( $p < 0.01$ ) related to ring-widths or BI-related indexed series. We built a linear regression (transfer-function model) based on those climatic variables which showed strong and temporally stable associations with the chronologies. Such transfer-function analysis is a linear regression model in which the tree-ring indexed variables (RW, EW, LW, dBI, EWBI, LWBI) were the predictors, and the climate-related factors temperature, precipitation, SPEI, VPD, NAO, SOI or WeMO were the predictands. Then, the skill of the selected proxy for reconstructing past climate was tested by performing a split-sample procedure for the most significant temperature correlation and was also used to verify model stability (Touchan et al., 2008). This procedure divides the full timespan covered by all records into two sub-periods (1925–1970 and

1971–2014), which were used as calibration and verification timespans, respectively. Furthermore, the reduction of error statistic (RE), and the Durbin–Watson test (DW) were computed to assess the suitability of the predictors for climate reconstructions beyond what is possible by simply using the calibration period mean of observed variable as the reconstruction (Biondi et al., 2008). The value of RE theoretically ranges from minus infinity to +1.0, and an RE value > 0 is considered positive skill of the model, and thus suitable for climate reconstructions (Fritts, 2001).

### 3. Results

#### 3.1. BI-based and RW-based variables differ in autocorrelation and sensitivity

Mean ring-width related values (RW, EW and LW) showed no significant differences among the study sites (Table 2). The mean ring width was found to be around 1 mm (time-series are presented in Fig. A1 of the Supplementary material). Also, statistical properties, like first order autocorrelation (AC) and correlation ( $\bar{r}$ ) statistics did not differ between ring width variables of the two black pine populations. Yet, at the low elevation site (LES) BI variables show a consistently lower AC than RW variables, which at the higher elevation site (HES) was found only for the dBI series (Table 2). The mean sensitivity is generally lower in BI as compared to RW variables, but slightly higher at LES (Table 2). The variance accounted by the first principal component is higher for RW variables on both sites (Table 2). Both at high and low elevation sites the RW, EW and LW series were strongly correlated with each other (Fig. A2), while correlations between BI series are less pronounced, although a strong correlation was found between dBI and LWBI, which is explained by the nature of the dBI series.

#### 3.2. Climate sensitivity

##### 3.2.1. Monthly level

Climatic signals captured by ring-width and BI related values are similar across sites and climatic factors, apart from the EWBI series at HES, which negatively reacted to previous December and seasonal winter temperatures (mean, maximum and minimum) (Fig. 2). The time-series of all variables at LES showed negative association with summer temperature, with LWBI and dBI containing stronger signal, which was not found at HES. On the other hand, at HES significant precipitation signal is observed in dBI, EWBI and LWBI, showing positive association between May precipitation and dBI, and negative – between September and May precipitation with dBI and EWBI, respectively. No significant correlation was found between ring size related variables and precipitation at HES. The opposite was found at LES, where strong significant positive seasonal (yearly, winter and autumn) precipitation effect on RWI, EWI and LWI, as well as on LWBI was observed (Fig. 2).

**Table 2**

Statistical characteristics of selected tree-ring chronologies from high- and low-elevation sites: tree-ring widths (RW, EW, LW) and blue intensity (dBI, EWBI, LWBI) for the common period 1900–2014. All dendrochronological statistics were calculated for standardized time-series except AC which was obtained from raw tree-ring width data. SD, standard deviation; AC, first-order autocorrelation for raw tree-ring series; Rbt, mean between-trees correlation of indexed values; MS, mean sensitivity; PC1, variance accounted for by the first principal component; EPS, expressed population signal.

Raw data	Low-elevation (LES)						High-elevation (HES)					
RW ± SD (mm)	0.95 ± 0.31						0.97 ± 0.29					
EW ± SD (mm)	0.64 ± 0.19						0.64 ± 0.17					
LW ± SD (mm)	0.30 ± 0.13						0.31 ± 0.12					
Residual chronologies	Low-elevation (LES)						High-elevation (HES)					
	RWI	EWI	LWI	dBI	EWBI	LWBI	RWI	EWI	LWI	dBI	EWBI	LWBI
No. trees	14	14	14	14	14	14	11	11	11	11	11	11
AC	0.60	0.60	0.47	0.40	0.51	0.42	0.64	0.66	0.45	0.38	0.65	0.58
$\bar{r}$	0.44	0.39	0.46	0.23	0.13	0.26	0.46	0.41	0.40	0.21	0.08	0.24
MS	0.30	0.29	0.46	0.28	0.03	0.06	0.26	0.24	0.45	0.28	0.02	0.03
PC1 (%)	51.13	46.23	52.38	37.33	19.97	34.37	54.56	49.02	48.54	36.79	30.84	43.19
EPS	0.91	0.90	0.92	0.81	0.68	0.83	0.90	0.88	0.88	0.75	0.48	0.77

##### 3.2.2. Daily level

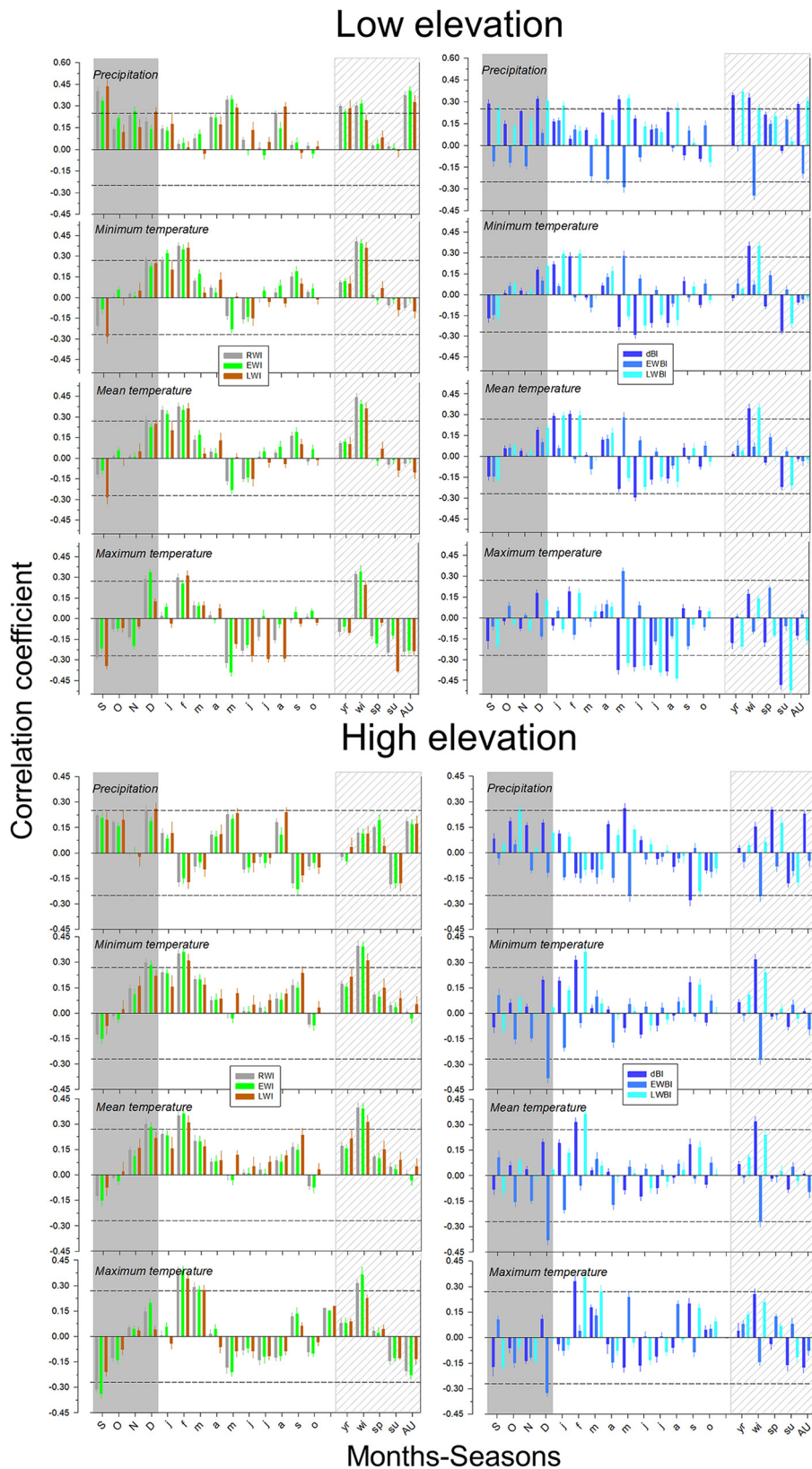
Daily climate-growth relationship analyses revealed that BI related variables of LES contain stronger mean temperature signal, compared to RW related series (Fig. 3). This difference is especially pronounced for dBI and LWBI series, where the correlation with summer temperature is exceeding −0.4 (Fig. 3b and f), whereas for RWI or LWI it is considerably lower (Fig. 3a and e). On the other hand, EWI series capture better an end-of-the-year and begin-of-the-year temperature signal (Fig. 3c). A similar pattern with stronger negative climatic signal in BI series was also found for maximum temperature (negative correlation with summer values), but the opposite – stronger signal in RWI, EWI and LWI than in dBI, LWBI and EWBI – was found for the minimum temperature (positive correlation with winter – early spring values, Figs. A3 and A4, respectively). While the signal strength of precipitation did not vary across the measured variables, we observed strong positive associations between the previous late autumn and the current summer for almost all the measured variables (Fig. A5). An exception was the EWBI series, where a negative effect of the spring precipitation was found (Fig. A5d). In contrast, at HES, the RWI, EWI and LWI were found to capture the temperature signal (mean, maximum and minimum) better than dBI, EWBI and LWBI series, showing a strong positive association with winter temperatures (Figs. A6, A7 and A8). However, dBI and LWBI series were found to contain a slightly stronger (negative) precipitation signal of the end of the growing season than RW series (Fig. 4b and f).

#### 3.3. Drought associations with growth and blue intensity

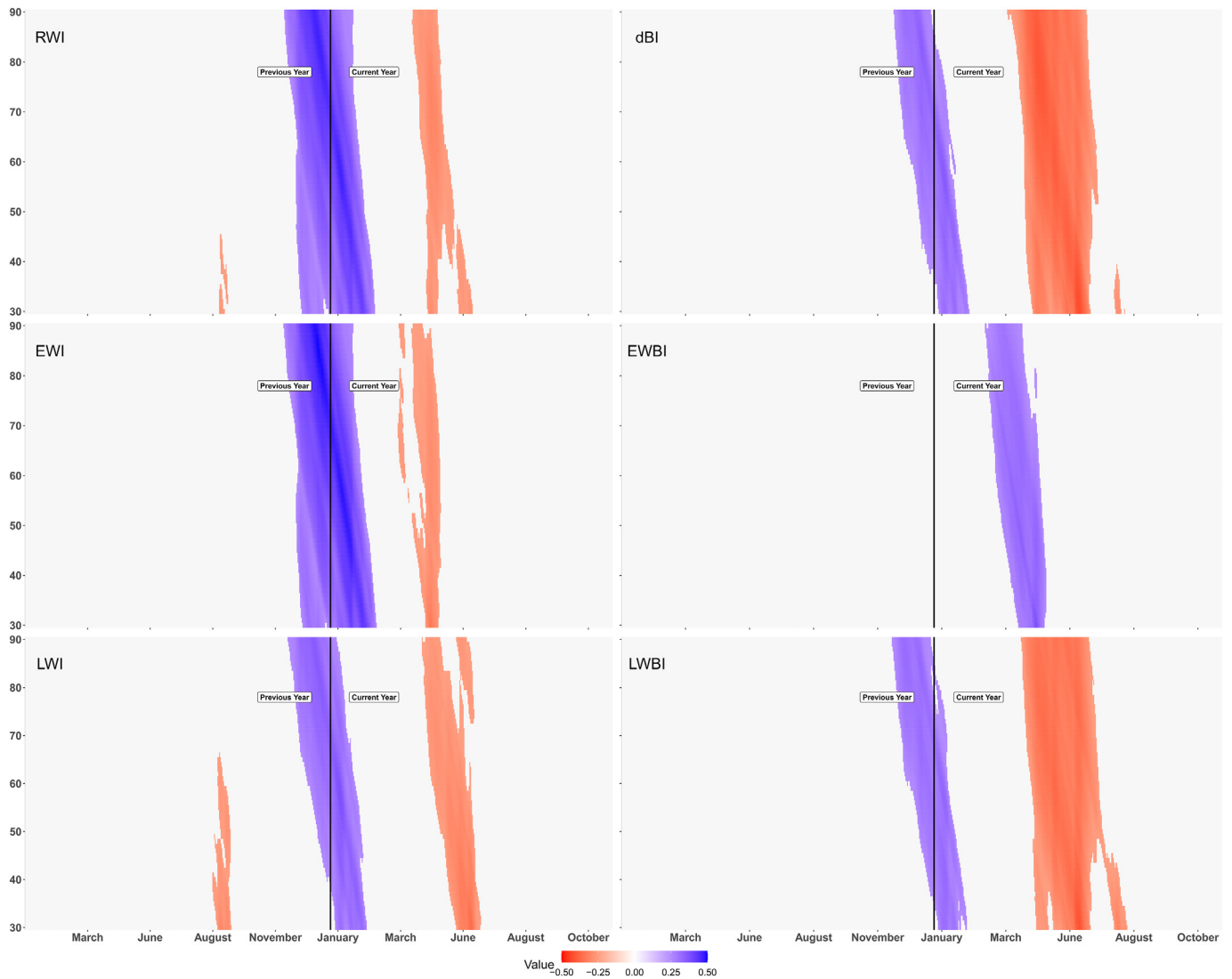
The correlation analyses between measured ring variables and SPEI showed a significant positive association of RWI, EWI and LWI series of LES and accumulative summer SPEI values recorded at 10–13 months long scales (Fig. 5a, left panels). A stronger correlation was observed between LES dBI and LWBI series with 14–15 months long scale SPEI (Fig. 5a, right panels). However, EWBI from LES was found to have negative association with early spring – summer SPEI of 9–10 months long scale. In contrast, at HES, RWI, EWI and LWI series showed stronger correlations with SPEI, than dBI, EWBI and LWBI (Fig. 5b). HES ring-width related series were positively associated with winter SPEI values of 5 to 15 months long scale (Fig. 5b, left panels).

#### 3.4. Sensitivity to atmospheric patterns

Few significant correlations were found between atmospheric circulation patterns (NAO, and SOI), WeMo and VPD with RW related variables (Fig. 6). Only monthly winter VPD was found to be significantly positively associated with EWI and RWI (Fig. 6, lower panels), both at LES and HES and EWI with monthly winter NAO at HES. Among the BI series, dBI was



**Fig. 2.** Bootstrapped Pearson's correlation coefficient ( $r$ ) for the relationship between the ring width indices (RWI, EWI, LWI), blue intensity indices (dBI, EWBI, LWBI) and the monthly climate variables (minimum, mean and maximum temperature and precipitation) for the common period 1950–2014. Each bar shows the mean correlation  $\pm$  standard error. The analyzed temporal window spans from previous year (uppercase letters) up to current growing season (lowercase letters). The grey vertical box includes the previous year and grey vertical area with line pattern shows annual (yr) seasonal (wi, winter; sp., spring; su, summer and AU, previous autumn) responses. Dashed horizontal lines show the 0.05 significance level.



**Fig. 3.** Bootstrapped Pearson's correlation coefficient ( $r$ ) for the relationship between the ring width indices (left column), blue intensity indices (right column) of LES and the mean daily temperature for the common period 1950–2014. The colors show the temporal patterns of climate-growth relationship for a specific window width of 90 days. For more results, see Supplementary Material, Fig. A3.

found to have significant negative correlation with seasonal winter and January SOI values at HES, and positive correlation with monthly VPD values at LES. Moreover, EWBI was found to be significantly negatively correlated with previous December VPD and positively with February NAO and HES.

### 3.5. Field correlations

For the field correlation analyses only factors with the largest difference in associations with the climate between RWI-related and dBI series, namely 12-months SPEI for LES was chosen. Performed analyses confirmed the existence of a stronger 12-months SPEI signal in LES dBI series (Fig. 7). The strongest correlation was found between LES dBI and the drought index, with the value  $> 0.6$  exactly matching the sampling area (Figs. 7b and 1a). Field correlations between precipitation and HES time-series did not reveal strong differences in climatic signals among the time-series.

### 3.6. Extreme droughts and resilience

The resilience, recovery, and resistance analyses of the derived series to the severe drought events (1995, 1999 and 2005) revealed overall higher resistance values of LWBI and EWBI series at both elevations (Fig. 8c and

d, right panels). The recovery, at the same time, was found to be higher in dBI series for all years, apart from year 2005 at HES, where high late-wood recovery values were observed (Fig. 8c and d, left panels).

Overtime, the growth resistance of the RW series was declining from 1995 to 2005 at both sites (Fig. 8a and b, right panels), while the recovery values presented an opposite trend for most of the variables increasing with time (Fig. 8a, b, c and d, left panels). The highest resilience values at the same time were observed during the 1999 drought events both at LES and HES (Fig. 8a and b, middle panels).

Regarding the density stability to severe droughts, no difference among the EWBI and LWBI resistance values over several dry years was observed, apart from the 2005 drought at HES, where higher EWBI resistance values were found (Fig. 8c and d, right panel). The dBI series, at the same time, demonstrated a decline trend in resistance values over drought events (Fig. 8c and d, right panels). The recovery values of the BI series demonstrated similar to RW series trend, with an increase from 1995 to 2005 (Fig. 8c and ds).

### 3.7. Potential of BI for drought reconstructions

Test on the potential of use of BI series for drought index (SPEI12) reconstructions revealed that LES dBI series have stronger power and positive



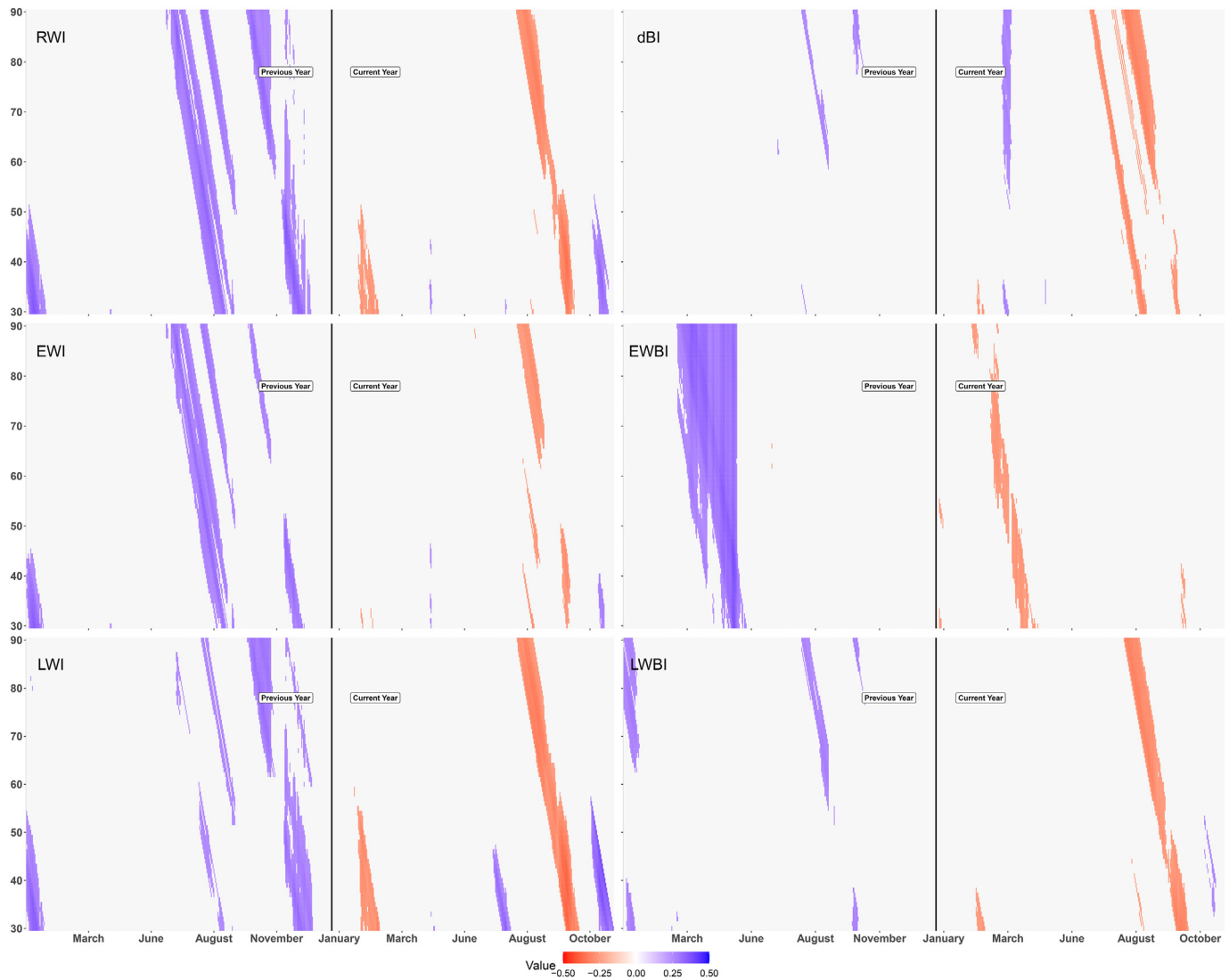


Fig. 4. Bootstrapped Pearson's correlation coefficient ( $r$ ) for the relationship between the ring width indices (left column), blue intensity indices (right column) of HES and the daily precipitation for the common period 1950–2014. The colours show the temporal patterns of climate-growth relationship for a specific window width of 90 days. For more results, see Supplementary Material, Fig. A4.

RE values (Table 3). RWI series, on the other hand showed less statistical power and some discrepancies between the measured and reconstructed series (Fig. A9b). The dBI series showed better reconstructing performance with mismatches between the reconstructed and measured values only for the beginning of the 20th century (Fig. A9a). The LWI and LWBI series have also shown poor skills for reconstruction of previous September and average current summer maximum temperatures, although the RE of the LWI was found to be higher, compared to LWBI (Table 3). Previous December minimum temperature was selected to test for the reconstruction skills test of EW and EWBI series derived from HES trees, and EWBI series have been found to have stronger skills, compared to EW series (Table 3).

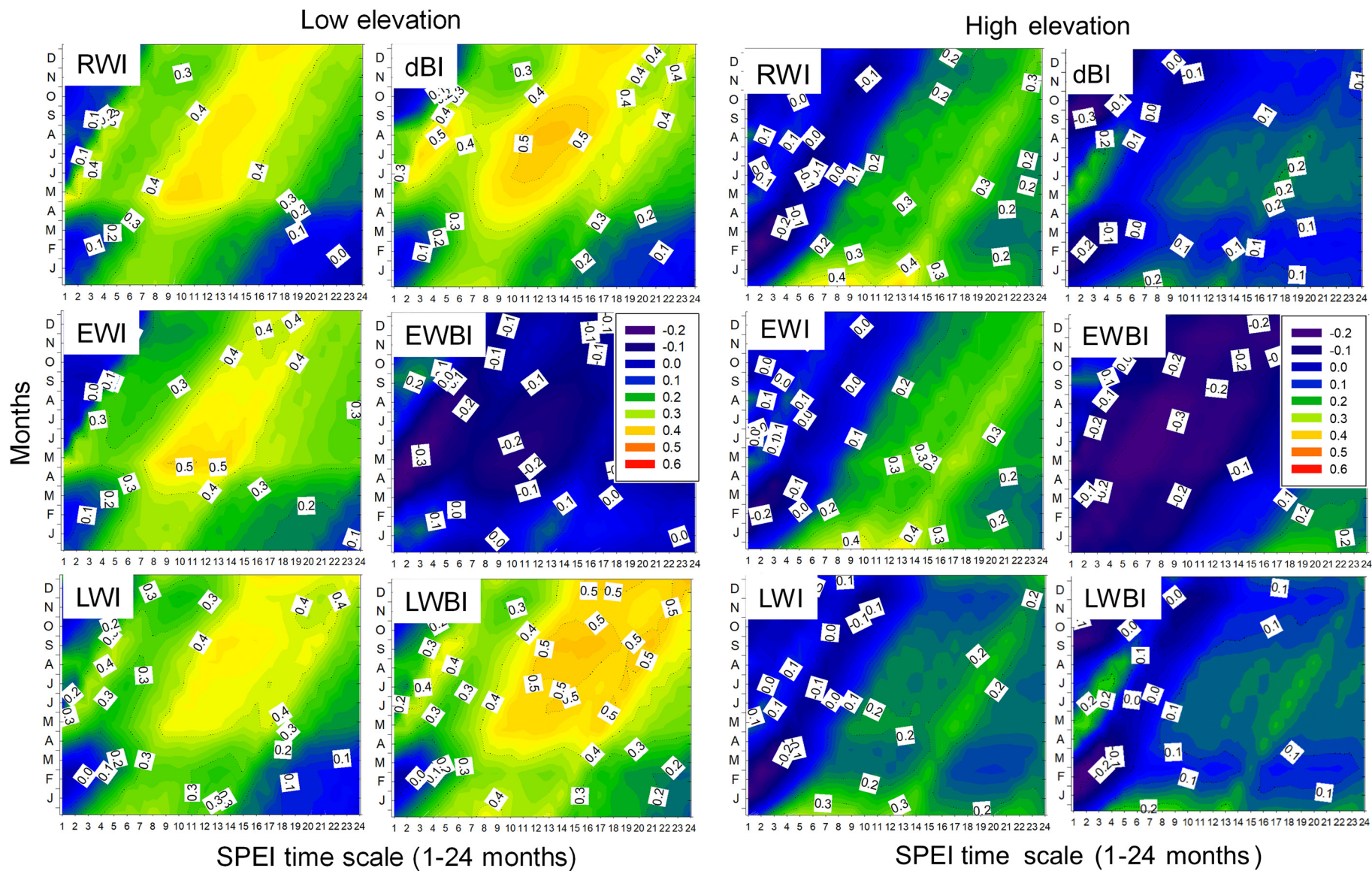
#### 4. Discussion

To our knowledge, this is the first study analyzing the potential of blue intensity variables for their applications in dendroclimatic or dendroecological research under Mediterranean conditions. Previous studies exploring the usefulness of the BI data were mainly carried out in higher latitudes with temperature being the strongest growth limiting factor (Björklund et al., 2014; Seftigen et al., 2020; Wang et al., 2020). Under the presented growing conditions, BI series have been found to be a better

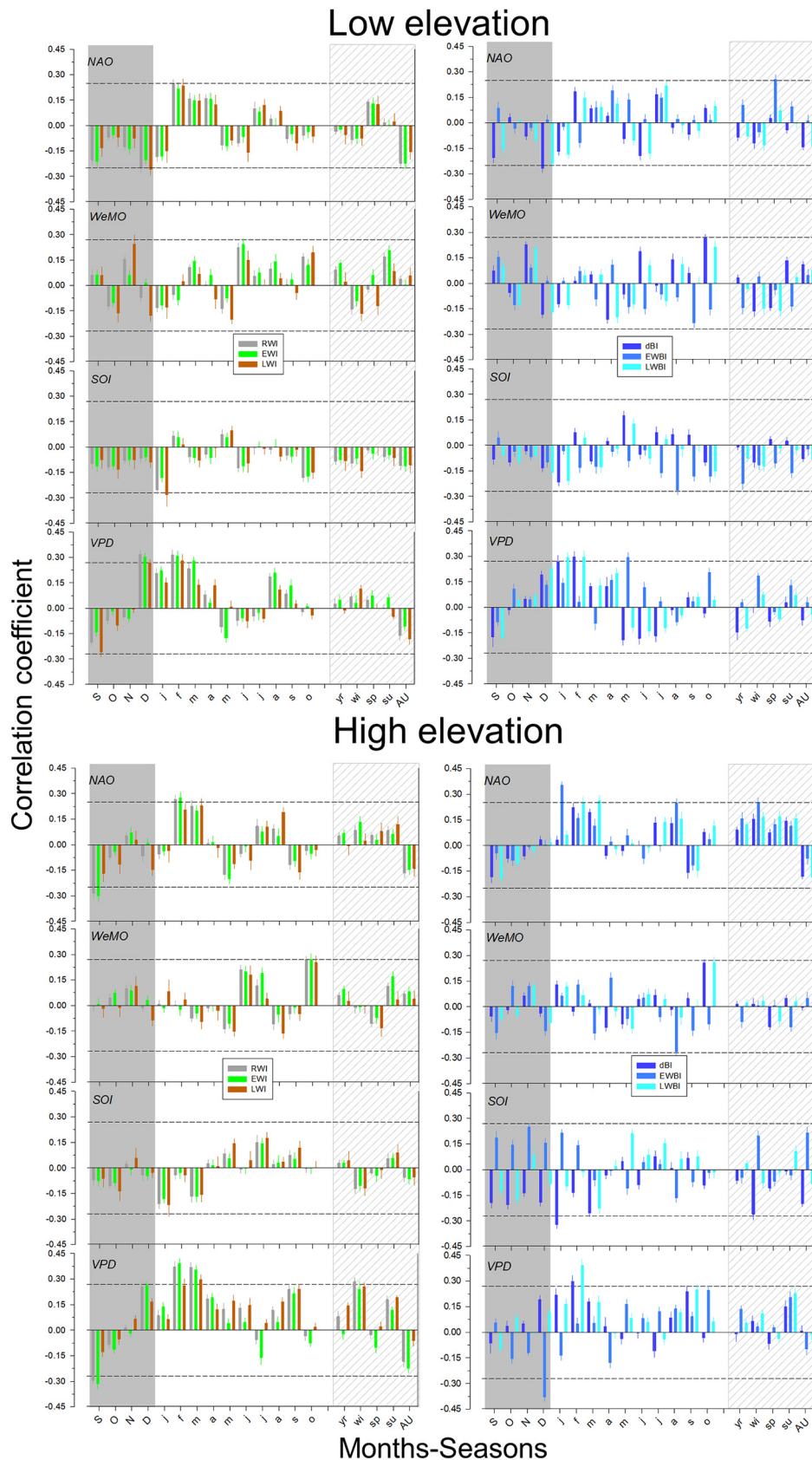
proxy than conventional tree-ring series for dendroclimatic analyses, especially for populations at lower elevations (Figs. 2–7). Moreover, a high potential of the BI data for use in ecological studies of conifers under drought-limited conditions was also discovered (Fig. 8).

##### 4.1. Blue as a proxy for better understanding climate effect on Mediterranean forests

Climate-growth relationships revealed differences both in climatic signal type and intensity between the measured BI and RW related variables, and this difference was found to be more pronounced at LES. The BI series can be interpreted as density values of the tree ring, due to strong similarities (Campbell et al., 2007) or sometimes indistinguishability among both (Wang et al., 2020), and are related to the relationship between cell wall thickness to the cell (tracheid) size (Björklund et al., 2021). With the smaller cell lumina and thicker wall (i.e., higher density), the water conductance efficiency is decreasing, but the mechanical strength and resistance to hydraulic failure is enhanced (Hacke et al., 2006; Pellizzari et al., 2016). Thus, at LES the BI series are directly linked to the xylem adjustments to growing conditions, and are thus more sensitive to environmental changes, which is demonstrated by the stronger climatic signal found at the site (Figs. 2 and 3). This discovered stronger response of the BI series



**Fig. 5.** Associations between drought index (SPEI) and standardized ring widths and blue intensity chronologies responses of *Pinus nigra*. The colour scale shows the correlations (Pearson coefficients) calculated between standardized series and the SPEI drought index obtained for 1- to 24-month-long scales (x-axes). The correlations were calculated for the common and best-replicated period 1950–2014 and considering the year of tree-ring formation (t). Correlation values above +0.20 and below –0.20 are significant at the  $P < 0.05$  level.



**Fig. 6.** Bootstrapped Pearson's correlation coefficient ( $r$ ) for the relationship between the ring width indices (RW, EW, LW), blue intensity indices (dBI, EWBI, LWBI) and circulation atmospheric patterns (NAO, North Atlantic Oscillation; WeMO, Western Mediterranean Oscillation; SOI, South Oscillation Index) and water deficit using Vapour Pressure Deficit (VPD) for the common period 1950–2014. Each bar shows the mean correlation  $\pm$  standard error. The analyzed temporal window spans from previous year (uppercase letters) up to current growing season (lowercase letters). The grey vertical box includes the previous year and grey vertical area with line pattern shows annual (yr) seasonal (wi, winter; sp., spring; su, summer and AU, previous autumn) responses. Dashed horizontal lines show the 0.05 significance level.

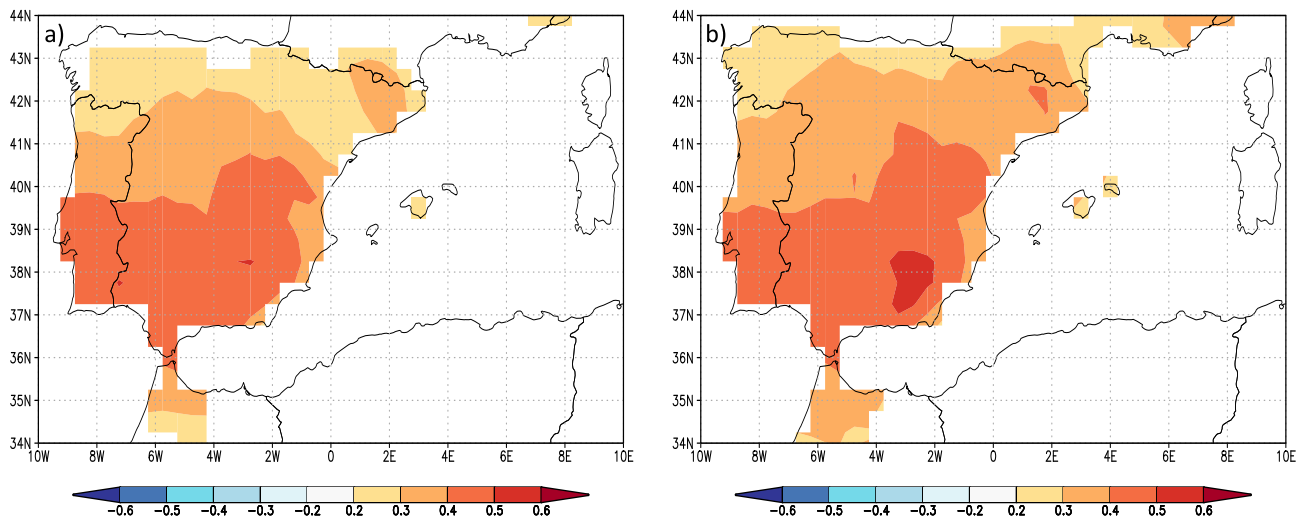


Fig. 7. Field correlations calculated between SPEI 12 months long scale and a) RWI and b) dBI indexed chronologies at LES.

(i.e., hydraulic properties) of the *P. nigra* to climate should allow a better understanding of the decline and mortality of the conifers in drought-prone regions by fast and efficient production of the proxy directly linked to early signals of drought induced mortality (López et al., 2021).

The signals captured by the RWI related series both at LES and HES (positive previous autumn precipitation and temperature together with positive winter temperature) were previously found in *P. nigra* ring-width chronologies from the area (Domínguez-Delmás et al., 2013) and in *P. canariensis* from Canary Islands (Rozas et al., 2013). The temperature signals can be explained as a reaction to change of the vegetation period with the warmer winters resulting in earlier start of the growing processes and resulting in extended growing season (Lebourgeois, 2000). The positive previous autumn precipitation signal found at LES RWI series is in line with the previous studies on conifers under Mediterranean conditions (Andreu et al., 2007; Camarero et al., 2015, 2021; Pasho et al., 2012) underlining the importance of autumn water for the species. Strong negative effect of the summer maximum temperature on RWI series from LES

is probably linked to warm-induced drought, so called “hotter droughts”, resulting in narrower rings (Pasho et al., 2012). This effect was found only at LES population, whereas HES did not reveal any negative associations with maximum temperature, probably due to the lower drought severities at the high-elevation site (Dorado Liñán et al., 2012). This was also confirmed with the SPEI – growth relationship analyses, where strong positive associations were found between summer SPEI 12 (accumulated SPEI index over 12 months) and LES series, whereas at HES the correlation values were considerably lower (Fig. 5), which might be triggered by difference in elevations, since sites from higher elevation were found to experience less droughts, compared to lower elevations (Sánchez-Salguero et al., 2015). The negative effect of droughts on growth of conifers is a common phenomenon in the region, and was often found in previous studies, explained by lower photosynthetic activity due to stomata closure as a consequence of the lower soil water availability (Medrano et al., 2002), and finally resulting in the production of narrow tree rings. (Camarero et al., 2013; Forner et al., 2018; Sánchez-Salguero et al., 2012). As

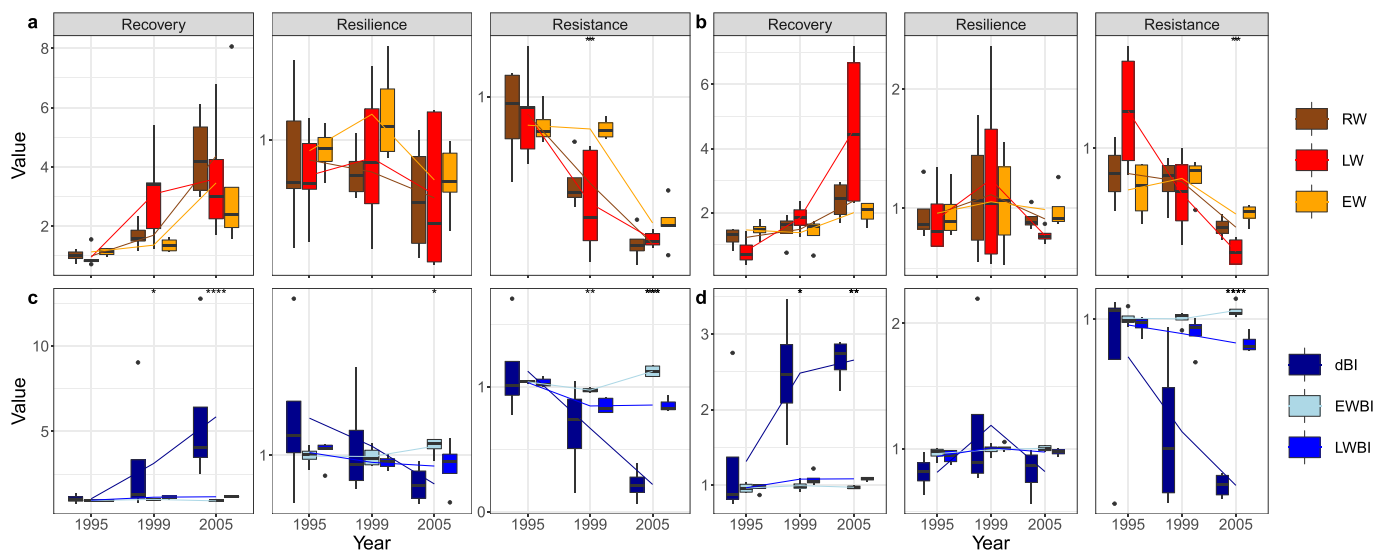


Fig. 8. Boxplots showing ring growth and latewood blue intensity stability during the extreme drought years (1995, 1999 and 2005) for a) LES RW, b) HES RW, c) LES BI and d) HES BI related series. Lines indicate trend in change of the mean values of the corresponding variable. Different stars indicate significant difference levels ( $p < 0.05, 0.01, 0.001$ ) between measured variables. In the box plots, the boundary of the box indicates the 25th percentile, a black solid line within the box marks the median, a dotted black line within the box indicates the 75th percentile. Whiskers above and below the box indicate the 10th and 90th percentiles. Points above and below the whiskers indicate outliers outside the 10th and 90th percentiles.

**Table 3**

Calibration-verification statistics for the dendroclimatic reconstructions of climatic variables using tree-ring widths and blue intensity variables for the period 1925–2014.

	Site	Calibration period	Verification period	RE	DW	RMSE	Correlation
RWI vs. SPEI12	LES	1971–2014	1925–1970	0.152	1.697	0.581	0.44
EWI vs. $T_{ID-1}$	HES	1971–2014	1925–1970	-0.052	2.549	1.236	0.28
LWI vs. $T_{Xs-1}$	LES	1971–2014	1925–1970	0.102	1.359	0.079	-0.30
dBI vs. SPEI12	LES	1971–2014	1925–1970	0.216	1.467	0.442	0.50
EWBI vs. $T_{ID-1}$	HES	1971–2014	1925–1970	0.139	2.247	1.166	-0.38
LWBI vs. $T_{Xsu}$	LES	1971–2014	1925–1970	0.080	1.469	1.116	-0.48

mentioned above, the LWBI and dBI of LES were found to have stronger associations with maximum temperature and drought index, compared to ring width related variables (Figs. 2 and 5) due to lower disturbance by other than climate external factors (Rydval et al., 2015). The spatial correlation analyses (Fig. 7) highlights benefit of using BI series due to the match of the strongest correlation with the sampling area. Thus, the use of BI series would allow for more direct assessment of the effect of the predicted climate change with increase in intensity and severity of drought events on the Mediterranean region. The positive (negative) associations between LWBI and dBI series with drought index (maximum temperature) had been previously found (with maximum density) for two pine species in Mexico (Pompa-García et al., 2021). The formation of latewood cells with wide lumen area and thin walls under water stress was linked to a shortage of non-structural carbohydrates (Medrano et al., 2002; Olano et al., 2014), while higher density (BI) values might result from enhanced carbohydrate production and rate of tracheid lignification due to wet summers (Galván et al., 2015). On the other hand, other studies mention that it is the duration of cell enlargement, and not the duration and rate of cell wall thickening mainly contribute to changes in cell wall thickening (Cuny et al., 2014). Decrease in MXD values as a result of intensive drought events and as a response to low soil water availability was also previously found in Scots pine stands in Northern Spain (Candel-Pérez et al., 2018) and related to production of less dense latewood tracheids for increase in water-conducting efficiency (Eilmann et al., 2011). In addition, under the Mediterranean conditions with high inter-annual climate variability there is a high chance of occurrence of intra-annual density fluctuation (IADF) as a result of more favorable growing conditions, e.g., precipitation events, in the end of summer/early autumn (Campelo et al., 2013). Consequently, IADFs will result in a decreased latewood density (LWBI). However, no IADFs were observed in the analyzed samples. It is noteworthy the unique climatic signal captured by the EWBI series (negative previous December temperatures and winter precipitation at HES; and negative (positive) with winter precipitation (May temperatures) at LES) (Fig. 2). The negative effect of overall positive for secondary growth climatic conditions on EWBI (earlywood density) was also found in the study of two Mexican pine populations (Pompa-García et al., 2021) and several conifer species from Spain (Camarero and Hevia, 2020), which can be explained by the formation of narrower cells under unfavorable conditions, leading to increase in density.

On the daily level, the climate – growth relationship assessment revealed an additional signal, which was not found on the monthly analyses - strong negative summer mean and maximum temperature association with the LES RWI, LW, EW, dBI and LWBI series (Fig. 3), which was probably masked due to the limitations of monthly climate – growth relationship analyses (Jevšenak, 2019), suggesting the use of daily analyses for more robust calculations of climate effect on tree growth. This limiting effect of summer temperatures on conifers growth under the Mediterranean conditions at LES might be result of the warming-induced drought. Again, the negative summer temperature effect was stronger at the BI related variables, highlighting importance of utilization of this variable in dendroclimatic studies in drought-prone regions.

In contrast, at HES, ring-width related variables were found to have stronger association with winter temperatures compared to BI series, though still low. Low correlations of the measured variables with the

climatic factors suggests either that there may not be one prevailing growth limiting factor at HES (Domínguez-Delmás et al., 2013; Dorado Liñán et al., 2012; Sánchez-Salguero et al., 2015). Moreover, local microsite conditions (e.g., soil, exposition, microclimate), which are very important as highlighted before (Düthorn et al., 2013), may hinder the results of the analyses at HES.

Complex local climate of Cazorla mountains (Domínguez-Delmás et al., 2013) might also be the reason for the low number of significant correlations with atmospheric circulation patterns (Fig. 6). However, BI values have demonstrated good potential for recording water stress signal at low elevations, and it can be extended for other drought-prone mountainous areas and to the analysis of drought induced dieback processes in areas with complex local climate systems and double stress (cold winters and hot summers with opposite – positive and negative – effects (Gea-Izquierdo et al., 2011; Martín-Benito et al., 2010)) hampering drought signals in ring-width series. Important to mention the cumulative nature of the SPEI index (SPEI 12 months) captured by the BI series, since long term water availability might be more important than summer precipitation under future climate conditions (Piovesan et al., 2008).

It is important to stress that in this study gridded climate data were used for calculations of climate-growth relationships, which also might obscure the effect of climate factors on the measured variables and limit deeper understanding of growth limiting factors in the area.

#### 4.2. Reconstruction potential of drought using blue series

The increase of drought-induced diebacks and reduction of vigor of conifer trees at global scale (Allen et al., 2015) and specially in drought-prone Mediterranean forests (Camarero et al., 2018) shows both an importance of understanding past climate variability and developing of a reliable proxy for drought reconstructions and forecasting (Sánchez-Salguero et al., 2017). Utilization of the latewood width values has shown a strong skill for summer drought reconstruction in Northeastern Spain (Tejedor et al., 2017). However, the maximum latewood values derived from *Pinus nigra* in Cazorla were selected for use in temperature reconstruction, and not drought (Esper et al., 2020), same as ring-width series (Dorado Liñán et al., 2015). In our study we have demonstrated potential of BI series for 12-months accumulated drought index (SPEI-12) reconstruction (Fig. A9 and Table 3). Further, the reconstruction skills statistics can be improved by using more trees for chronology construction, as minimum 25–30 trees were found to be needed to ensure high signal quality in BI series (Wilson et al., 2019) or choosing different detrending methods, e.g., regional standardization curve, which has shown good potential in the same area (Esper et al., 2020). Despite these limitations, the potential of BI series for drought reconstruction might have strong implications for mountainous Mediterranean conifer forests.

#### 4.3. Blue for ecology

The similarity between density and BI series opens new possibilities for utilization of BI series in dendroecological studies, especially considering the importance of the density containing the first signals of forest decline (López et al., 2021). This study was the first attempt to use BI series for resilience analyses. The stronger resistance of EWBI and LWBI values to major drought events in the area (Fig. 8), which is also reflected by a lack of variability in the resilience and recovery indices of the variables, indicates that trees, despite producing a lower number of cells (i.e., narrower rings), try to keep the proportion between the cell wall and cell size stable during the major drought events and thus to maintain hydraulic capacity of the ring (López et al., 2021). These findings require further investigations on the BI series potential for ecological studies involving several tree species growing under contrasting climatic conditions. For example, the first attempt to link BI with defoliation events of larch trees has also shown promising results (Arbellay et al., 2018), and also demonstrate great possibilities of the utilization of BI series to assess effects of insect outbreak events, e.g., bark beetles in Central Europe, which is one of the major forest declines problems in the area

(Hlásny et al., 2021), or other major disturbances events. The latter is especially relevant for reconstruction of the past forest management activities and opens new perspectives for dendro-historical studies.

## 5. Conclusions

Our study shows that blue intensity series are suitable proxies for dendroclimatic studies of conifer trees under the Mediterranean conditions. Stronger associations with temperature, and especially with the drought index at low elevation, open possibilities to increase our understanding of future climate change impact in hotspots of forest decline and forest loss areas. Since BI reflect processes related to wood formation and hydraulic behavior, we propose that blue intensity series can be integrated in long-term reconstruction of past climate variability in Mediterranean regions. Our findings also indicate that the use of daily climate series incorporates a stronger signal linked to growing season conditions than classical monthly series.

## CRedit authorship contribution statement

**Linar Akhmetzyanov:** Conceptualization, Methodology, Data collection, Original draft preparation.

**Raúl Sánchez-Salguero:** Conceptualization, Methodology, Supervision, Reviewing and Editing.

**Ignacio García-González:** Reviewing and Editing.

**Marta Domínguez-Delmás:** Reviewing and Editing.

**Ute Sass-Klaassen:** Reviewing and Editing.

## Data availability

Data will be made available on request.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.scitotenv.2022.159291>.

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