Tree Physiology 35, 1378–1387
doi:10.1093/treephys/tpv085

Research paper

Divergent climate response on hydraulic-related xylem anatomical traits of *Picea abies* along a 900-m altitudinal gradient

Daniele Castagneri[1,2], Giai Petit[1] and Marco Carrer[1]

[1]Università degli Studi di Padova, Dept Territorio e Sistemi Agro-Forestali, Viale dell'Università 16, I-35020 Legnaro (PD), Italy; [2]Corresponding author (daniele.castagneri@unipd.it)

Received May 15, 2015; accepted August 5, 2015; published online September 15, 2015; handling Editor Maurizio Mencuccini

Climate change can induce substantial modifications in xylem structure and water transport capacity of trees exposed to environmental constraints. To elucidate mechanisms of xylem plasticity in response to climate, we retrospectively analysed different cell anatomical parameters over tree-ring series in Norway spruce (*Picea abies* L. Karst.). We sampled 24 trees along an altitudinal gradient (1200, 1600 and 2100 m above sea level, a.s.l.) and processed 2335 ± 1809 cells per ring. Time series for median cell lumen area (MCA), cell number (CN), tree-ring width (RW) and tree-ring-specific hydraulic conductivity (Kr) were crossed with daily temperature and precipitation records (1926–2011) to identify climate influence on xylem anatomical traits. Higher Kr at the low elevation site was due to higher MCA and CN. These variables were related to different aspects of intra-seasonal climatic variability under different environmental conditions, with MCA being more sensitive to summer precipitation. Winter precipitation (snow) benefited most parameters in all the sites. Descending the gradient, sensitivity of xylem features to summer climate shifted mostly from temperature to precipitation. In the context of climate change, our results indicate that higher summer temperatures at high elevations will benefit cell production and xylem hydraulic efficiency, whereas reduced water availability at lower elevations could negatively affect tracheids enlargement and thus stem capacity to transport water.

Keywords: conductivity, lumen area, Norway spruce, tracheid, tree ring.

Introduction

Xylem is responsible for water and solutes transport from the root tips to the leaves and for providing mechanical support in all the vascular plants (Tyree and Ewers 1991, Růžička et al. 2015). In conifers, both these functions are ensured by a single type of cells, the tracheids. In cold and temperate environments, such cells are formed by the cambium (secondary growth) over a defined period, the growing season, forming distinct annual rings (Fritts 1976). The number and characteristics of tracheids produced yearly vary over time according to internal and external tree factors. For example, the number of cell rows, which mainly determines radial growth rates (Mäkinen et al. 2003, Dufour and Morin 2013), usually follows a decreasing size–age trend in trees growing in an open canopy environment without significant competition constraints (Cook and Kairiukstis 1990). On the other side, a consistent increasing pattern is commonly observed in the mean cell lumen size from the stem pith outwards, more pronounced in the so-called juvenile wood (Carrer et al. 2015). This reflects the ontogenetic increase of xylem conduits, required to stabilize total hydraulic efficiency as the tree becomes taller with age, widely observed in different plant species and environments (West et al. 1999, Anfodillo et al. 2006).

Despite these universal age–size patterns, environmental changes can influence the process of xylem formation (xylogenesis), determining intra- and inter-annual variability in the number and size of xylem cells, and therefore altering the stem capacity to transport water and to sustain the tree (Wilkinson et al. 2015). In a climate change context, increasing the mechanistic understanding of climate influence on tree xylem structure

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is, therefore, critical to improve model predictions of terrestrial ecosystem responses to future climatic conditions (Fonti et al. 2010, Camarero et al. 2015). In this sense, intensive monitoring of cambial activity during the growing season can shed light on the environmental effect on xylogenesis phases (Rossi et al. 2006, Lupi et al. 2010, Cuny et al. 2014). However, the limit of this approach is the temporal length, with data spanning mostly a few (generally 1–5) years, whereas climate change is inherently a long-term process covering time frames from decades to thousands of years. It is, therefore, crucial to also investigate longer timescales, to correlate the time dimension of xylem plasticity with the investigated environmental trends (Moreno-Gutiérrez et al. 2012).

In this sense, retrospective analysis of xylem anatomical features along tree-ring series can certainly provide the required long-term perspective. However, most past studies have investigated climate influence on specific cell features rather than focusing on the corresponding year-to-year variability in the whole ring structure coupled with the related consequences for tree physiology and growth (Olano et al. 2012). Such studies evidenced different, and in some cases contrary, cell anatomical responses to climate. For example, water availability has been reported to either favour (Martin-Benito et al. 2013, Pritzkow et al. 2014) or reduce (Eilmann et al. 2009) lumen diameter of earlywood tracheids in *Pinus sylvestris* (L.). Similarly, high summer temperature can have a positive (Fonti et al. 2013, Liang et al. 2013) or negative effect (Bryukhanova and Fonti 2013, Xu et al. 2013, Szymczak et al. 2014) on cell size of different conifers. Inconsistency in results is likely related to site-mediated growth responses, an aspect scarcely investigated in conifer wood-anatomy studies so far.

Whatever the sample design and species under investigation, most previous studies had to tackle several methodological constraints, such as the subjective selection of few (usually five) cell rows within a ring (Vaganov et al. 2009, Fonti et al. 2013). This approach cannot provide information on the whole tree-ring anatomical structure, i.e., on the total number and size distribution of cells, critical information for an assessment of the conductivity of tree-ring structure (González-González et al. 2014, Oladi et al. 2014, Wilkinson et al. 2015). As water transport in conifers is ensured by the rings forming the sapwood (Mencuccini 2003), investigation on inter-annual climate forcing on the whole tracheid complex in tree rings would allow long-term climate influence on xylem functioning to be assessed.

The objective of this study is to increase the mechanistic understanding of tree xylem response to climate under different environmental conditions, with a specific focus on anatomical traits that determine stem radial growth and regulate hydraulic function. Applying recently enhanced methods of sample preparation and methodological advances in image processing (von Arx and Carrer 2014), we aimed at analysing all the cells within a ring section (two orders of magnitude larger) over longer time spans compared with past studies (Carrer et al. 2015). Our goal was to get more reliable inferences on the association between xylem traits and climate, to assess the link between xylem anatomical variables and stem hydraulic functioning.

The investigation was performed on 24 *Picea abies* (L.) Karst. trees along a 900-m altitudinal gradient that represents an ideal setting to investigate species plasticity, i.e., the capacity of a relatively homogeneous genetic population to acclimate to different environmental conditions (Piotti et al. 2009, Chevin et al. 2013). Using cell chronologies from three sites at 1200, 1600 and 2100 m above sea level (a.s.l.), and daily-resolved climate records, we evaluated (i) how climate variability differentially influences cell lumen area and cell number (CN) along the altitudinal gradient and (ii) how the variability of these two anatomical traits affects tree-ring width (RW) and tree-ring-specific hydraulic conductivity.

Materials and methods

Study area and sample collection

The study area is located on a north-east facing slope above Cortina d'Ampezzo in the Eastern Italian Alps (46°30′N, 12°07′E). Daily maximum temperature averages 20.8 °C during July and 3.1 °C in January. Mean annual precipitation, occurring as snow during wintertime, is 1080 mm, with a maximum in June (125 mm) (Cortina d'Ampezzo meteorological station, 1230 m a.s.l., 1926–2011). Soils are shallow and calcareous, and forest stands have a predominance of Norway spruce from the valley bottom to the tree limit at 2150 m a.s.l., mixed with other coniferous species like *Larix decidua* Mill., *Pinus cembra* L., *P. sylvestris* and *Abies alba* Mill.

Three study plots were located on the same slope at 1200 (EL12), 1600 (EL16) and 2100 (EL21) m a.s.l. at a distance of ~1 km from one another. Considering the typical lapse rate of 0.6 °C/100 m of the lower troposphere (Körner 2007), our transect covered a temperature gradient of ~5 °C. Between 2011 and 2013 at each site, two 5-mm increment cores were extracted at breast height from 15 to 20 dominant and codominant Norway spruce trees; tree height and diameter at breast height were also recorded.

Cell measurement along tree-ring series

Ring widths were measured to the nearest 0.01 mm using TsapWin (Rinntech, Heidelberg, Germany), and cross-dating accuracy of ring-width series was checked using COFECHA (Holmes 1983). For cell analysis, we selected one core per tree for a total of 24 cores (eight for each site) among those closer to the pith and without visible faults such as nodes, reaction wood, or rotten or missing parts.

Cores were divided into 4–5 cm pieces, and transversal sections (15–20 μm thick) were prepared with a rotary microtome (Leica, Heidelberg, Germany), stained with safranin (1% in

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distilled water) and permanently fixed with Eukitt (BiOptica, Milan, Italy). Digital images were captured with a light microscope at ×40 magnification (Nikon Eclipse 80) and stitched together with PTGui (New House Internet Service B.V., Rotterdam, The Netherlands). The images (Figure 1a) were then processed with ROXAS (version 1.6, von Arx and Carrer 2014). Anatomical analyses were performed on the whole tree-ring series, so that 5162 rings with an average of 2335 ± 1809 cells were processed. For each ring, we assessed (i) median cell lumen area (MCA), (ii) total CN, (iii) RW and (iv) tree-ring-specific hydraulic conductivity (Kr), i.e., the sum of hydraulic potential conductivity of all the cells (Kh), in an area with fixed tangential width of 1 mm and a variable radial length corresponding to the ring width (González-González et al. 2014). Kh for each cell was automatically calculated by ROXAS as approximated by Poiseuille's law and adjusted to elliptical tubes (Nonweiler 1975), according to the formula:

$$Kh = \frac{\rho \cdot CA \cdot m}{\eta \cdot k}$$

where ρ is the density of water, CA is cell area, m is the mean hydraulic radius calculated for elliptical section tubes, η is viscosity of water and k is a coefficient depending on the eccentricity of the ellipse.

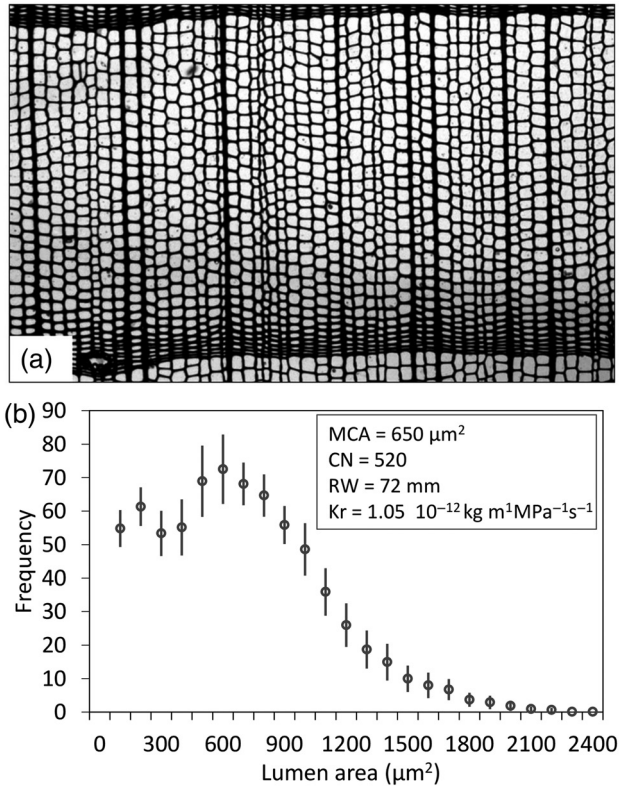

Figure 1. Image of the 2008 ring section of a *P. abies* sample at EL16 (a) and cell lumen area distribution (mean frequency and standard errors on 100 µm² classes, scaled on 1 mm width image) of all the sampled trees in the same area and year (b). The corresponding MCA, total CN, RW and tree-ring-specific hydraulic conductivity (Kr) are reported in the inset.

To assess how MCA and CN (explanatory variables) affected RW and Kr (response variables), we implemented mixed linear models, with site as a random factor. In addition, we used partial correlation analysis to estimate the relationship between an explanatory variable and a response variable after controlling for the effects of the other predictor (Cohen et al. 2013). Models were implemented using Rcommander (Fox 2005), lme4 (Bates et al. 2014) and pcor (Kim and Yi 2007) packages in R (R Development Core Team 2014). Variables were log transformed to achieve normal distribution.

Tree ring and cell chronologies

In order to study the climate effects on the inter-annual variability in tree rings and their anatomical characteristics, we built time series (chronologies) for each parameter (MCA, CN, RW and Kr). To remove age–size-related trends, usually present not only in ring-width series but also in lumen-area series (Carrer et al. 2015), we applied a two-step standardization procedure. Individual series were first standardized by fitting a stiff function (power function or 200 years cubic spline with 50% frequency cut-off) to raw data series and dividing observed by expected values. Then, to emphasize high-frequency variability, these dimensionless indices were submitted to a second detrending procedure fitting a cubic smoothing spline with 50% frequency cut-off of 20 years and we computed the observed vs expected ratio a second time. Autoregressive modelling was used to remove the serial autocorrelation (Cook and Kairiukstis 1990) to better emphasize the high-frequency domain contained in the time series.

At plot level, mean chronologies for each parameter were calculated as bi-weight robust mean of the detrended individual series (Cook 1985). Some statistics have been considered to compare the different time series: the mean sensitivity (MS), to estimate the year-to-year variability (Fritts 1976); the first-order autocorrelation [AC(1)], to measure the influence of previous year's growth on the following year and the mean correlation between trees (R_{bar}), to appreciate the common signal among trees. Chronology elaborations were performed with detrendeR (Campelo et al. 2012) R package.

Climate–growth relationships

We estimated the climate effect on tree-ring and anatomical traits by using daily records of maximum temperature and precipitation from 1926 to 2011 obtained from the meteorological station of Cortina d'Ampezzo, 1230 m a.s.l., located at ~3 km from the study plots. A daily climate data set allows better detection of the short-term climate influence on cell parameters by avoiding the rough and artificial aggregation in 12 months (Dufour and Morin 2013, Fonti et al. 2013). We investigated the relation between tree-ring and anatomical variables with climate data by

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using CLIMTREG V.4 (Beck et al. 2013, Liang et al. 2013). This software sums up (precipitation) or averages (temperature) daily meteorological data at different time windows ranging from 21 to 121 days. Moving correlations with xylem variables were then calculated for all the time windows from previous year 1 July to 30 October of the current ring formation year for the period 1927–2011.

Linear models to assess the combined effect of different climate parameters (explanatory variables) on the four chronologies (MCA, CN, RW and Kr response variables) in the three plots were then computed. The climate parameters identified by CLIMTREG were submitted to a stepwise selection process, using Bayes–Schwarz information criterion to select the final model composed just of significant variables and taking into account the frequent multicollinearity among climate parameters. This method is suitable for models with a limited number of predictors (Aho et al. 2014) and is more restrictive in the variables selection than other customary methods (e.g., Akaike information criterion). All the final models were tested for linearity of functional relations (Ramsey test), absence of residual autocorrelation (Durbin–Watson test), normality of residuals (Shapiro–Wilk test) and reduced inter-correlation of the explanatory variables (variance inflation factor and correlation matrix). The models were implemented using Rcommander (Fox 2005) and ppcor (Kim and Yi 2007) packages in R.

Results

Tree-ring and cell characteristics along the altitudinal gradient

Trees at low elevation (EL12) were taller and younger than those at intermediate (EL16) and high (EL21) elevation (Table 1) and presented a different tree-ring anatomical structure. Despite the similar frequency distribution of tracheid lumen areas in all the plots (i.e., with many small- to medium-size tracheids and a long tail of large earlywood tracheids, Figure 1b), at EL12, trees formed more and larger cells per year ($P < 0.001$, Kruskal–Wallis median test). This led not only to nearly double the mean RW but also significantly increased the tree-ring-specific conductivity (Kr) with respect to higher elevation sites (Table 1).

Considering the complete data set (24 trees and 5157 rings), RW was mainly related to CN, with just a slight positive influence of MCA at EL16 and EL21 (Figure 2, Table 2). Instead, Kr was the result of the combined effect of CN and MCA.

Regarding the mean chronologies (see Figure SA1 available as Supplementary Data at *Tree Physiology* Online), MS and R_{bar} were generally higher for RW, Kr and CN compared with MCA (Table 1), which had a higher first-order autocorrelation. For all the parameters, autocorrelation was higher at EL16, while MS and R_{bar} were generally higher at EL21. Correlations of MCA with CN (and RW) chronologies were low (Table 3), especially in the lower plots ($r = 0.40$ at EL21, not significant at EL16 and EL12), suggesting different environmental drivers for CN with respect to cell lumen area.

Climate–growth models

Climate–growth models performed better at high than at low elevation (Figure 3, Table 4, Table SA1 available as Supplementary Data at *Tree Physiology* Online). Their performances were fairly good for Kr at high and intermediate elevation, but models underestimated positive and negative peaks at EL12 (Figure 3). Moving along the altitudinal gradient, the most important climate driver for tree-ring and xylem anatomical traits shifted from temperature at EL21 to precipitation at EL12 (Figure 4). Temperature and precipitation influence are hereafter described according to three seasonal groups: (i) summer of the year prior to ring formation, (ii) previous winter and spring and (iii) current growing season.

Table 1. Characteristics of sampled trees, xylem parameters and chronologies in the three study plots EL21, EL16 and EL12. Samples, height, diameter and years (maximum age) refer to the trees. Unit and median refer to the four xylem parameters, i.e., MCA, total CN, RW and tree-ring-specific hydraulic conductivity (Kr). Different letters indicate significantly different median values according to Kruskal–Wallis (K–W) test. MS, AC(1) and R_{bar} are mean sensitivity, first-order autocorrelation and mean correlation between trees included in the mean chronologies.

Plot	Trees				Xylem parameters				Chronologies		
	Samples	Height	Diameter	Years	Parameter	Unit	Median	K–W	MS	AC(1)	R_{bar}
EL21	8	20 m	55 cm	235	MCA	μm^2	490	a	0.080	0.860	0.266
					CN	–	820	a	0.182	0.764	0.381
					RW	mm	0.869	a	0.184	0.708	0.444
					Kr	kg m MPa^{-1} s^{-1}	0.863×10^{-12}	a	0.231	0.741	0.416
EL16	8	30 m	57 cm	286	MCA	μm^2	492	a	0.068	0.946	0.186
					CN	–	856	a	0.161	0.870	0.244
					RW	mm	0.910	a	0.162	0.899	0.289
					Kr	kg m MPa^{-1} s^{-1}	0.927×10^{-12}	a	0.200	0.890	0.283
EL12	8	37 m	69 cm	124	MCA	μm^2	721	b	0.080	0.789	0.191
					CN	–	1357	b	0.160	0.783	0.192
					RW	mm	2.066	b	0.144	0.784	0.273
					Kr	kg m MPa^{-1} s^{-1}	2.416×10^{-12}	b	0.193	0.760	0.199

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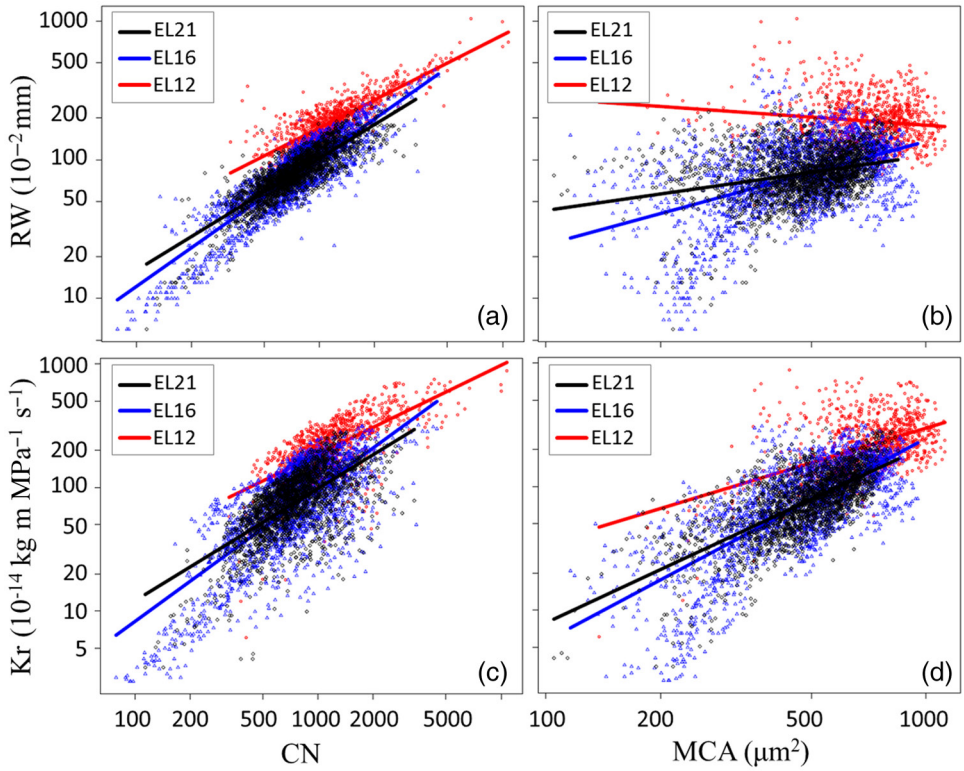

Figure 2. Relationships between RW and CN (a), RW and MCA (b), Kr and CN (c), and Kr and MCA (d) in the three study sites EL21, EL16 and EL12. Axes are log scaled.

Table 2. Parameters of the linear models for RW and Kr as a function of MCA and CN. The squared semi-partial correlation coefficient (S.s.cor.) represents the variance explained by a predictor residualized with respect to all the other predictors. ***$P < 0.001$, n.s., not significant.

	Estimate	SE	*t*-value	S.s.cor.	R^2_{adj}	Sig.
RW model					0.923	***
(Intercept)	−4.489	0.058	−77			
MCA	0.577	0.007	78	0.15		***
CN	0.861	0.005	184	0.66		***
EL16						***
EL21						***
Kr model					0.999	***
(Intercept)	−11.740	0.007	−1682			
MCA	1.529	0.001	1395	0.42		***
CN	0.999	0.001	1386	0.45		***
EL16						n.s.
EL21						n.s.

Table 3. Correlations between MCA, CN, RW and Kr mean chronologies within the three plots. **$P < 0.01$, ***$P < 0.001$, n.s., not significant.

	CN	Kr	RW
EL21			
MCA	0.40***	0.63***	0.51***
CN		0.85***	0.82***
Kr			0.78***
EL16			
MCA	$0.14^{n.s.}$	0.36***	0.30**
CN		0.87***	0.80***
Kr			0.82***
EL12			
MCA	$0.13^{n.s.}$	0.51***	$0.06^{n.s.}$
CN		0.84***	0.69***
Kr			0.64***

Previous year summer emerged as a key season significantly affecting inter-annual variability of most of the parameters (in 11 out of 12 models, Figure 4, Table SA1 available as Supplementary Data at *Tree Physiology* Online). At the highest elevation (EL21), MCA, CN and RW were negatively affected by temperature and no evidence was detected of significant correlations with precipitation. A negative effect of temperature was found for RW and CN in trees at intermediate elevation (EL16), whereas precipitation significantly stimulated MCA and Kr. At the lowest site (EL12), a positive effect of precipitations emerged for MCA, CN, RW and Kr, with an additional negative effect of temperature on RW (Figure 4, Table SA1 available as Supplementary Data at *Tree Physiology* Online).

The period just after the end of the previous growing season (October–November) seemed barely important for the next-year growth along the whole gradient with the exception of a moderate effect of temperature on CN at EL16 (Figure 4). On the contrary, winter precipitation, mostly occurring as snow, was one of the most consistent and positive influences on radial growth and xylem traits at all the sites (10 out of 12 models). At EL21, precipitation after late January positively affected radial increment

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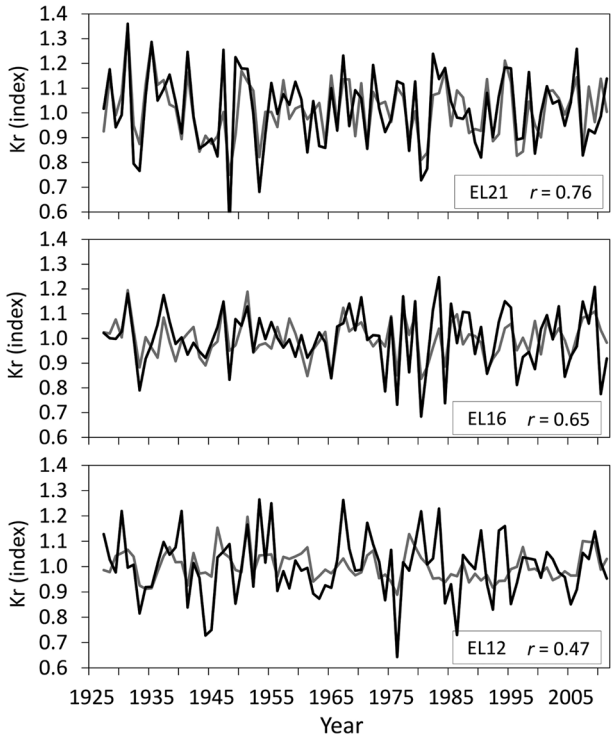

Figure 3. Visual comparison between measured (black) and predicted (grey) Kr chronologies at EL21, EL16 and EL12. Pearson's *r* between measured and predicted chronologies is reported in the insets.

Table 4. R^2_{adj} of the 12 models computed to assess the climate influence on tree-ring and cell features in the three plots. All the models are significant at $P < 0.001$.

	MCA	CN	RW	Kr
EL21	0.591	0.540	0.524	0.554
EL16	0.399	0.480	0.418	0.384
EL12	0.346	0.163	0.391	0.187

and tree-ring anatomical traits. At EL16, precipitation between January and March affected CN, RW and Kr, while at lower elevations (EL12), MCA, RW and Kr were favoured by December to February precipitation. Temperature effect over this period had a less clear pattern. Cold temperature between March and early May had a positive effect on all parameters at the highest elevation (EL21), with the April–May period being of particular importance for MCA and, therefore, for Kr. At a lower elevation (EL16), a slight negative effect of early April temperature was recorded for RW and, even with a lower intensity and over a shorter period, for Kr. Mild temperatures in January–February stimulate RW, but not the other variables, at EL12.

During tree-ring formation (early summer), the models evidenced divergent sensitivities along the altitudinal gradient (Figure 4). Temperature was the main actor at EL21: high early season temperatures (around June) strongly and positively affected all tree-ring parameters. At EL16, where the growing season is likely anticipated compared with EL21, temperature influence on CN, RW and Kr occurred earlier. At the lowest site (EL12), temperatures seemed to have negligible influence on tree-ring size and anatomy. In July, precipitations had a slight positive effect on MCA at EL21 and, in the late growing season, also on Kr. At mid-altitude (EL16), precipitation during most of the summer time benefited MCA, while precipitation towards the end of the growing season had a moderately negative influence on the other parameters. At EL12, June to early July precipitations benefitted MCA, RW and Kr, but not CN.

Discussion

Our study evidenced how quantitative tree-ring anatomy now allows a thorough assessment of inter-annual variability of xylem traits in conifers. The latest technical advances permit xylem features to be examined in detail with an adequate sample size in terms of tree and CNs, together with a longer time frame in line with the typical dendrochronological investigations. Thoroughly measuring the number and lumen area of all the cells within ring sections, we were able to reliably assess the tree-ring-specific hydraulic conductivity (Kr), a measure of the contribution of a given ring to the total stem potential conductance.

Our results showed that environmental conditions associated with the altitudinal gradient ultimately affect xylem structure and functioning of *P. abies*, inducing significant differences in the growth performances of the species. In agreement with investigations on other conifer species (Mäkinen et al. 2003, Eilmann et al. 2009, Olano et al. 2012, Dufour and Morin 2013, Martin-Benito et al. 2013), we showed that ring width in *P. abies* mainly depends on CN. Wider rings at EL12 were mostly due to higher CN compared with EL21 and EL16. Furthermore, trees at EL12 produced larger cells. In accordance with the axial conduit widening theory (West et al. 1999, Anfodillo et al. 2006), larger cells allow water transport to a longer pathway, and thus, trees can be taller. Our results indicate that a reduction in the cell lumen area (at breast height) in trees at high elevation (also observed on other species, Pourtahmasi et al. 2011, Olano et al. 2013), coupled with reduced CN, caused a reduction in rings specific hydraulic conductivity (Kr) and, consequently, on the whole stem conductance. Reduced water transport has negative effects for photosynthetic assimilation and growth of high-elevation trees (Petit et al. 2011, Körner 2015).

Both cell lumen size and number contributed to determine tree-ring-specific hydraulic conductivity. However, our analyses revealed that these two variables are, at least in some cases, differently affected by intra-annual climate variability. Although most current knowledge on climate influence on xylem characteristics in conifers comes from studies on xylogenesis, which usually ignore climate conditions prior to the growing season (Lugo et al. 2012), we demonstrated, from a longer-term perspective, that cell features can also be strongly influenced by previous growing season conditions. Climate of the previous

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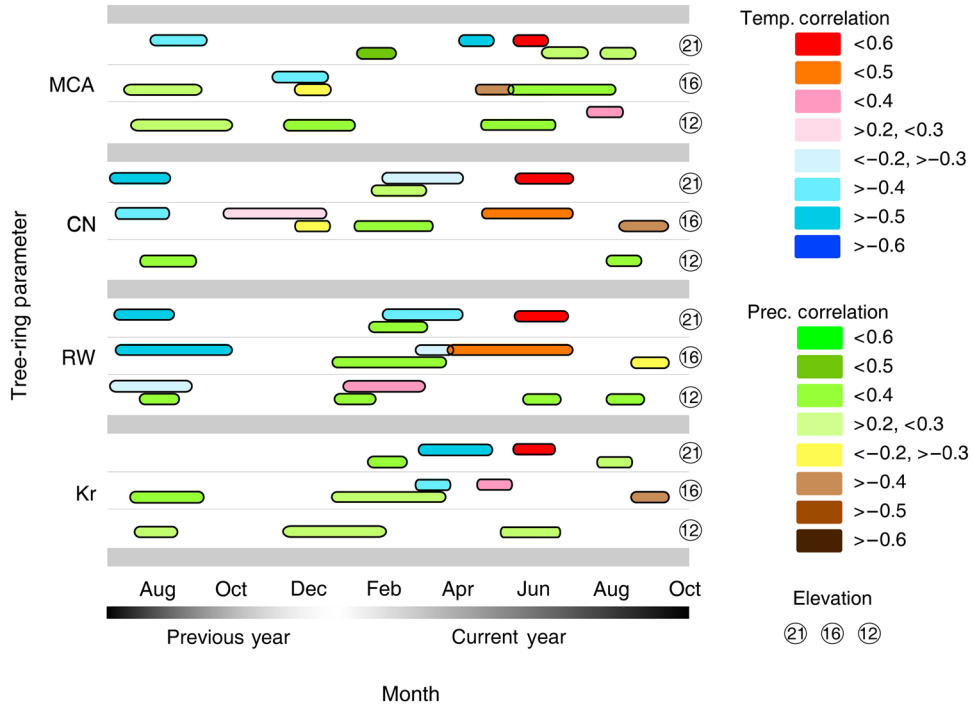

Figure 4. Graphical representation of the 12 models to assess temperature and precipitation influence (from previous year July to current year October) on MCA, CN, RW and Kr at EL21, EL16 and EL12. The bar type is related to the partial correlation coefficient (see key), while bar length indicates the period of significant correlation.

year likely influences carbon reserves for the following growing season (Xu et al. 2013, Palacio et al. 2014). In addition, along the whole altitudinal transect, winter precipitation had a positive effect on most xylem characteristics (i.e., it increased MCA, CN, RW and Kr). Negative effect of February–April temperature at EL21 and EL16 could be related to early snow melting (Kirdyanov et al. 2003). Snow cover shelters above- and below-ground tree tissues from extreme winter events (Oberhuber 2004, Lupi et al. 2010) and represents a fundamental water source for the following months (Duchesne et al. 2012, Olano et al. 2013). Given the importance of winter precipitation for tree growth, outcomes and inferences of studies on xylem dynamics and modelling could be improved by incorporating not just climate information of the current growing season, but also of several months prior to cambial reactivation.

Summer temperature represented the most limiting factor for tree growth at high elevation. In particular, CN showed the highest sensitivity to June–July temperatures, whereas no effect was found for temperatures in the period immediately preceding cambial reactivation. Our results, based on a time series of 85 years, hence support xylogenesis models that indicate higher summer temperatures as the key factor for higher annual cell production in cold environments (Fritts et al. 1991, Deslauriers et al. 2003, Rossi et al. 2006, Cuny et al. 2012), rather than an early onset of the growing season (Vaganov et al. 1994, Myneni et al. 1997, Dufour and Morin 2010, Lupi et al. 2010, Lugo et al. 2012, Xu et al. 2013). Furthermore, our analysis indicated that, at the species altitudinal limit, warm conditions during early summer are not only critical for cell production but also for cell enlargement. Close to the species altitudinal limit, low temperatures during the short growing season limit metabolic processes, such as the fixation of photosynthates (Körner 1998, Rossi et al. 2008, Petit et al. 2011), affecting xylogenesis in both the differentiation and enlargement phases. At lower elevation, we observed a gradual reduction of summer temperature influence on tree-ring anatomy, being negligible at EL12. Interestingly, at the intermediate elevation (EL16), temperature had a moderate influence on CN, but none on cell lumen area.

On the contrary, precipitation effect on tree-ring anatomy increased from high to low elevation. Despite a strong June temperature effect, a moderate positive effect of precipitation between July and early August emerged on cell lumen area at EL21, pointing out that water availability in the second part of the growing season can influence cell enlargement even at high elevations. At EL16 and EL12, high precipitation (in an early period at EL12) promoted larger cell lumen formation (but not higher CN). Cell enlargement is among the physiological processes more sensitive to water constraint (Taiz and Zeiger 2006), and a different effect of short-term water availability on cell enlargement and cell production has been observed in controlled experiments on tree seedlings (Abe et al. 2003). Here, we provide long-term empirical evidence of the different effect of inter-annual variability of water availability on cell enlargement (related to cell lumen area) and production (related to CN) in adult trees.

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The climate signal in tree-ring-specific conductivity (Kr), which depends on both CN and size, quantified the environmental forcing on the tree-ring potential to transport water. At high elevation (EL21), high summer temperature benefit on CN and size induced an increase in Kr. At lower elevation (EL16), Kr sensitivity to summer climate was somehow reduced by the balance between the divergent climate responses of CN, mainly positively affected by temperature, and cell lumen area, mostly related to precipitation. In warmer conditions, compared with the other sites (EL12), summer water availability started to play a key role in the xylem hydraulic complex, as evidenced by June and July precipitation benefit on Kr.

The research setting allowed us to speculate on future xylem response to climate warming of *P. abies*, one of the most drought-sensitive conifer species in Central Europe (Castagneri et al. 2014, Lévesque et al. 2014), under different environmental conditions. At the species altitudinal limit, enhanced summer temperature should induce higher radial growth rates, a more efficient water transport and, thus, taller trees (Lupi et al. 2010, Petit et al. 2011). All these aspects need be considered in growth and carbon sequestration models for high altitude and latitude forests (Hu et al. 2010, Smith and Dukes 2013). At mid-elevation, higher summer temperature could promote cell production and radial growth, but to a lesser extent. Although tree-ring potential conductivity seems not to be threatened by enhanced temperature at mid and low elevations, a future drier climate in the Southern Alps (Giorgi and Lionello 2008) could affect cell enlargement and induce lumen shrinkage, even under optimal environmental conditions for the species. This could compromise water supply to the upper part of the crown (Hentschel et al. 2014, Hereş et al. 2014), especially in tall trees with wide conduits, and potentially lead to forest decline, as already observed in many forests normally not considered at drought risk (Choat et al. 2012).

Supplementary data

Supplementary data for this article are available at *Tree Physiology* Online.

Acknowledgments

The authors are grateful to Vinicio Carraro, Enrico Marcolin, Francesco Nicolini, Elena Pellizzari and Angela Luisa Prendin for their help in laboratory work, Georg von Arx for support in using ROXAS software and Alison Garside for revising the English. This research is linked to activities and benefited from the fruitful discussions conducted within the COST FP1106 'STReESS' network.

Conflict of interest

None declared.

Funding

The study was supported by University of Padova (Research Project D320.PRGR13001, Senior Research Grants 2012).

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