



UNIVERSITÀ DEGLI STUDI DEL MOLISE

Dipartimento Agricoltura, Ambiente e Alimenti

Dottorato di ricerca in

Difesa e Qualità delle Produzioni Agro-Alimentari e Forestali

XXV ciclo

TESI

S.S.D. AGR/05

**“THE INFLUENCE OF LAND-USE AND CLIMATIC CHANGES ON MOUNTAIN PINE
(*PINUS MUGO* TURRA SPP. *MUGO*) ECOTONE DYNAMICS AT ITS SOUTHERN RANGE
MARGIN ON THE MAJELLA MASSIF, CENTRAL APENNINES.”**

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ANNO ACCADEMICO 2012/2013

ABSTRACT

Mountain pine *krummholz* within the Majella National Park forms an isolated patch and is a relictual species, thought representing the potential natural vegetation of the subalpine belt in Central Apennines, Italy. Mountain pine grows above the timberline and has in the Majella massif its southernmost limit of the distributional range. This vegetation dominates the treeline ecotone, between the dense beech forest and the alpine prairies, and is highly sensitive even to minor environmental changes. For this reason, it is a major proxy for monitoring the effects of climate and land-use changes occurring in the Mediterranean Basin. Mediterranean climatic trend includes projected increasing temperature and a different distribution of total precipitation, which are increasing during winter and decreasing during summer and autumn, respectively. Moreover, in Central Apennines, subalpine shrublands have been deeply reduced from the past anthropic practices. The recent abandonment of marginal areas and grazing cessation are allowing a recolonization of areas previously grazed.

The main objective of this study was to assess the effects of land-use and climatic changes on mountain pine population dynamics in the Majella massif, using three different methodological approaches. Temporal and spatial distribution of mountain pine since 1954 was reconstructed and a multitemporal map was created by historical aerial ortophotos. Results documented an upward and downward expansion of mountain pine and an ingrowth into the formerly tree-free grassland in early 1900s, together with a decline of the local human population and livestock.

The relative role of climatic variables on mountain pine growth was assessed by dendroclimatological analysis. Climate-growth relationships were assessed by correlation function (CF) analysis and moving CF (MCF), investigating the possible deviations in the response of mountain pine to recent climate change. Results demonstrated that climate variables affected mountain pine growth at the treeline ecotone in the Majella massif. Significant correlations were found with the spring maximum temperatures and summer precipitation. MCF showed that mountain pine is a sensitive species, in terms of tree ring growth, particularly at the beginning and at the end of the growing season.

For better understanding dendroclimatological results, wood anatomical analyses were conducted. Timings of mountain pine xylogenesis were assessed and the principal phenological phases (cambial division; enlargement; wall thickening; mature) were monitored during the entire growing season. At the same time timings of compression wood (CW) formation and possible differences with the opposite wood (OW) formation were investigated. Results indicated that CW is characterized by higher number of cells and thicker secondary wall, as well as longer xylogenesis compared with the OW. The growing season started simultaneously in both types of wood (on 20 May), and ended two weeks earlier in OW (on 22 September) than in CW (on 6 October). Increasing temperature of spring month would be favourable for the elongation of mountain pine growing season, which would

probably start early, though decreasing precipitation could compromise wood formation during summer, mountain pine being highly sensitive to this climatic parameter.

Growing above the limit of tree vegetation, mountain pine proved to be a promising species for monitoring climate- and land use-driven influences over tree-ring growth at high elevation. An excessive expansion of mountain pine *krummholz*, mainly at higher altitudes, may represent a threat for rare and endemic species growing in the alpine grasslands of the Majella National Park. However, conservation and preservation of mountain pine are required, in order to avoid a severe reduction of its distribution and then the risk of extinction of the last “island” present in the Apennine.

RIASSUNTO

La mugheta del Parco Nazionale della Majella è una formazione isolata e relittuale, testimonianza vivente della possibile serie vegetazionale climacica della fascia subalpina in Appennino centrale. Il pino mugo cresce solitamente sopra il limite altitudinale degli alberi (*timberline*) e trova nel massiccio della Majella il limite meridionale del suo areale di distribuzione. Questa vegetazione ecotonale, di transizione tra il bosco denso di faggio e le praterie alpine, sensibile anche alle minime variazioni ambientali, rappresenta un ottimo indicatore degli effetti dei recenti cambiamenti climatici e di uso del suolo in atto nel Bacino del Mediterraneo. Le tendenze climatiche previste per quest'area includono un aumento delle temperature ed una diversa stagionalità delle precipitazioni, con aumento nei mesi invernali e diminuzione nei mesi estivi e autunnali. In Appennino, inoltre, la vegetazione arbustiva del piano subalpino è stata drasticamente ridotta in passato dall'azione diretta dell'uomo, per far spazio al pascolo d'alta quota. L'attuale e progressivo abbandono dei pascoli d'alta quota, però, sta permettendo una ricolonizzazione del piano alpino con la libera evoluzione delle serie vegetazionali.

Questo lavoro ha l'obiettivo di valutare, mediante tre diversi approcci metodologici, gli effetti delle variazioni d'uso del suolo e dei cambiamenti climatici sulle dinamiche evolutive dei popolamenti di pino mugo nel massiccio della Majella. Un confronto diacronico svolto a scala di paesaggio, su fotogrammi aerei opportunamente ortorettificati e ortofoto digitali, ha mostrato una progressiva espansione della mugheta. Il contributo di indagini condotte in archivi di Stato e sedi ISTAT ha dimostrato una concomitante drastica riduzione della pratica del pascolo.

Il ruolo delle variabili climatiche nella crescita del pino mugo è stato indagato mediante analisi dendroclimatologiche su carote legnose di pino mugo. Le relazioni clima-accrescimento sono state definite mediante funzioni di correlazione (CF), la cui variabilità nel tempo in risposta ai cambiamenti climatici è stata valutata con funzioni di correlazione a finestra mobile (MCF). I risultati hanno dimostrato che le variabili climatiche influenzano la crescita del pino mugo nell'ecotono della *treeline* sul massiccio della Majella. Significative CF sono state ottenute con le temperature massime

primaverili e le precipitazioni estive. Le MCF hanno dimostrato che il pino mugo è una specie sensibile alle variazioni climatiche, in termini di crescita radiale, in particolare all'inizio e alla fine della stagione vegetativa. Il significato delle correlazioni ottenute è stato indagato con analisi anatomiche svolte a livello di singolo individuo che hanno permesso di definire le tempistiche della xilogenesi nel pino mugo. Le diverse fasi fenologiche (divisione e crescita cellulare, ispessimento della parete secondaria e completa lignificazione delle tracheidi) sono state monitorate durante un'intera stagione vegetativa, indagando contemporaneamente sulle tempistiche di formazione del legno di compressione (CW) e sulle differenze con il legno opposto (OW). Un maggior numero di cellule con parete secondaria più spessa è stato osservato nel CW, con tempi di formazione più lunghi rispetto al OW. Maggio rappresenta l'inizio della stagione vegetativa del pino mugo sul massiccio della Majella, settembre e ottobre la fine. Un incremento delle temperature primaverili permetterebbe un'anticipazione della riattivazione vegetativa e di conseguenza un allungamento della stagione di crescita del pino mugo.

È quindi possibile confermare un'evoluzione della mugheta dovuta soprattutto all'abbandono dei pascoli, ma anche influenzata dagli andamenti climatici. Una eccessiva espansione della mugheta, principalmente verso quote maggiori, rappresenterebbe una minaccia per le specie rare ed endemiche presenti nei pascoli alpini del Parco Nazionale della Majella. Tuttavia, politiche di conservazione e salvaguardia dovranno allo stesso tempo evitare una eccessiva riduzione della mugheta e quindi la scomparsa dell'ultimo nucleo presente in Appennino.

Ringraziamenti

Seppur breve e impegnativo questo dottorato mi ha permesso di conoscere persone umanamente e professionalmente straordinarie, collaborare con i migliori esperti del settore, vivere esperienze altamente formative ed ammirare meraviglie che solo la natura può creare. Molte persone durante questi anni mi hanno aiutato a migliorare e proseguire questo progetto di ricerca e ad ognuna di esse va il mio più grande ringraziamento. Ne elencherò molte ma sicuramente ne dimenticherò alcune, ad ogni modo non per importanza ma in ordine cronologico di “comparsa”!!

La figura professionale che sin dall’inizio mi ha seguita in questo lavoro, consigliandomi pur lasciandomi sempre “carta bianca” su qualsiasi decisione, è il Prof. Roberto Tognetti, Tutor del mio Dottorato di Ricerca. Lo ringrazio in particolar modo per avermi aiutata a portare avanti le mie idee.

A lui si aggiungono i docenti e i ricercatori dell’Università del Molise che, in ufficio o sul campo, sono stati fondamentali per la preparazione e realizzazione dei lavori svolti, grazie ai proff. Marco Marchetti e Gherardo Chirici e ai dott.ri Vittorio Garfi, Bruno Lasserre, Fabio Lombardi, Giovanni Pelino, Claudia Cocozza, Giovanni Santopuoli, Mauro Maesano e Lorenzo Sallustio.

Ringrazio ancora una volta il dr. Paolo Cherubini, per i suoi preziosi consigli e per avermi accolta nuovamente in una struttura tecnicamente perfetta quale il WSL di Birmensdorf. Con lui il dr. Patrick Fonti che mi ha seguita durante tutto il semestre svolto presso i laboratori del WSL, rendendo possibile la mia formazione nel campo dell’anatomia del legno. Ad essi devo aggiungere un’altra “esperta” in materia, la dr.ssa Giovanna Battipaglia, senza la quale mi troverei ancora a scrivere l’introduzione del lavoro! La ringrazio infinitamente per la tanta pazienza che ha avuto nei miei confronti e la continua disponibilità.

Ringrazio l’Ente Parco Nazionale della Majella e il Corpo Forestale dello Stato - Ufficio Territoriale per la Biodiversità di Pescara per le autorizzazioni concesse e per l’interesse attivo nei confronti di questo progetto di ricerca. In particolare desidero ringraziare la geologa Elena Liberatoscioli, per il suo indispensabile contributo nella parte cartografica del lavoro, ed i forestali Luciano Schiazza, Giuseppe D’ascanio, Gianni Blasioli e Roberto Iezzi che, accompagnandomi settimanalmente nell’area di studio, mi hanno permesso di svolgere agevolmente il lavoro di campo durante tutto il 2011. Inoltre, coadiuvati dal supporto di alcuni operai, hanno contribuito all’istallazione della stazione termometrica che ancor’oggi continua a registrare i dati di temperatura a 1900 m di altitudine. Grazie ad essi ho scoperto un territorio fantastico ed un ambiente di lavoro esemplare, basato esclusivamente sulla passione e sul senso del dovere.

Spunti, suggerimenti o semplici chiacchierate hanno sempre avuto un ruolo molto importante per lo svolgimento di questo lavoro e numerose sono le voci che ho avuto il piacere e l’onore di ascoltare. Tra i tanti, ringrazio il Prof. Hein van Gils, lo scrittore Edoardo Micati e gli esperti di anatomia Holger Gärtner and Fritz Schweingruber del WSL per i loro preziosi consigli.

Un ringraziamento speciale va alla mia famiglia che, nonostante le numerose difficoltà, non ha mai smesso di incoraggiarmi e di credere nelle mie capacità.

Infine, il mio più grande ringraziamento va ad Eugenio che mi sopporta pazientemente giorno dopo giorno, spingendomi ad essere sempre ottimista e intraprendente, e che, inconsapevolmente, ha contribuito alla realizzazione del cuore di questo lavoro in cambio di fantastiche avventure sulla nostra "Montagna Madre".

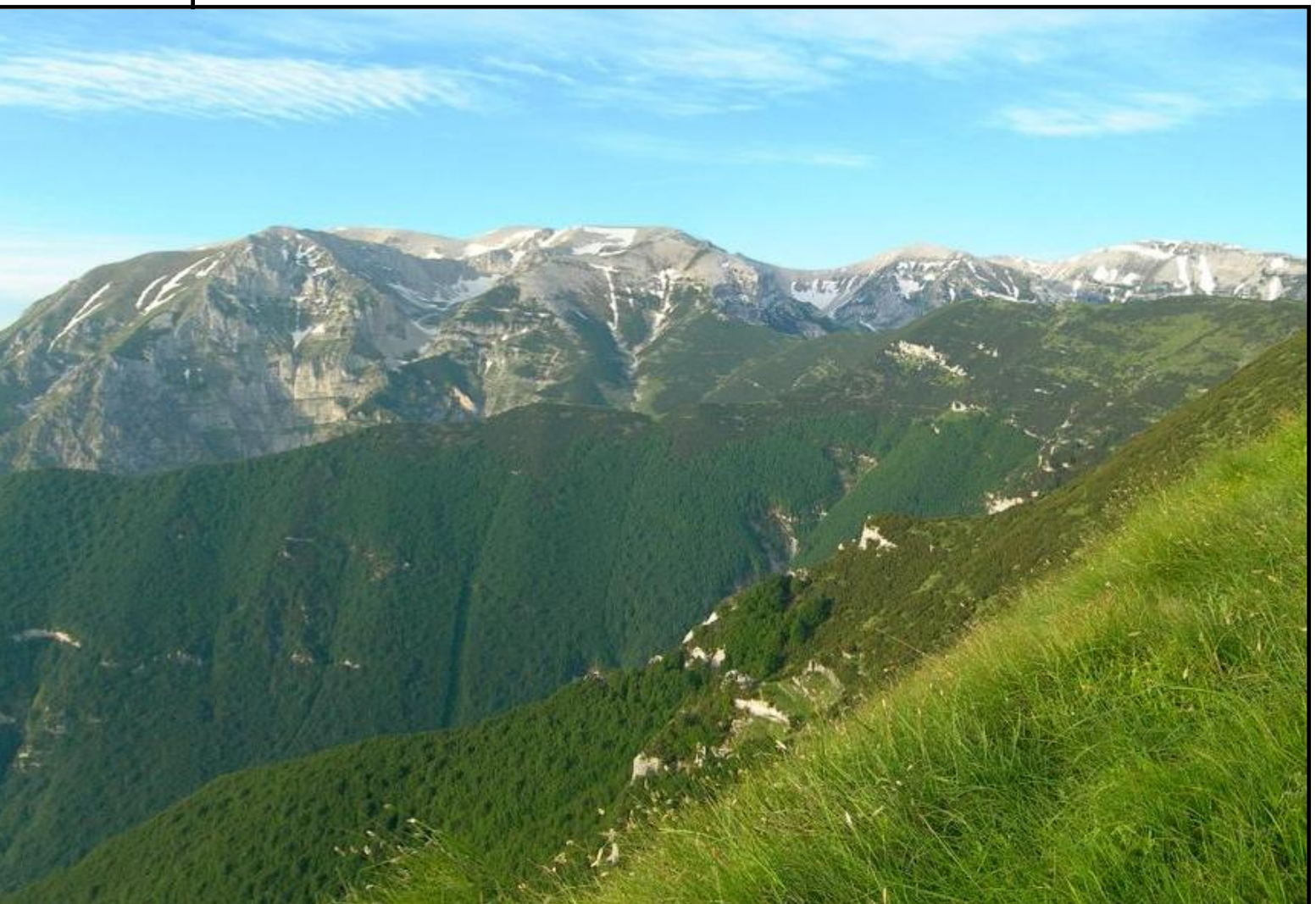
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FIRST CHAPTER:

General introduction



1.1 Global changes in the Mediterranean mountain ecosystems

The Mediterranean Basin is a very sensitive ecosystem to global climatic variations. Given its location in a transitional zone between the arid climate of North Africa and the temperate and rainy climate of central Europe, the Mediterranean climate is more variable and vulnerable also to minor changes in atmospheric dynamics (Lionello et al. 2006). Despite its restricted distribution, this area is considered as one of the global hotspots of biodiversity, hosting about 25000 plant species, 50% of which are endemic (Médail and Quézel 1999; Myers et al. 2000). In addition, in Mediterranean mountain systems, direct and indirect effects of land use changes are particularly significant due to long-standing human impact on the original landscape heterogeneity, and to the intense changes in structural and functional features for the majority of forests (Marchetti et al. 2010).

The high biodiversity and the climate variability of Mediterranean basin are strictly correlated to the great percentage of mountain areas present in its territory. A recent book, focusing on Mediterranean mountain environments (Vogiatzakis 2012), pointed at the interaction between human and physical processes as responsible for the continuum change in this environment. Metzger et al. (2005) recognized Mediterranean mountains as separate entities influenced by both the Mediterranean zone they are situated in, and a distinct mountain climate. Mediterranean reliefs are affected by erosion processes more than by glacial abrasion, in comparison with other European mountains, and receive more precipitation than the surrounding lowlands, being sources of various important rivers (Vogiatzakis 2012). They were defined as the backbone of the whole region, including the largest islands, occupying more than 50% of land in many Mediterranean countries (Regato and Salman 2008). These environments usually display distinct variations in climate and associated biological phenomena from their base to the summit (Price 1981). The diverse topography and altitudinal range resulted in a variety of microclimates and environments along a short distance, often with sharp transitions (ecotone) in vegetation sequences. As a consequence, mountains exhibit high biodiversity and a high number of endemic species, because many species remained isolated at high elevations. Some endemics are relictual, since Mediterranean mountains acted as glacial refugia for many European plant and animal species, providing suitable habitats for many taxa of the Northern Hemisphere and being the sources for post-glacial recolonization, when temperatures rose again at the end of the glacial period (Médail and Diadema 2009). Other endemics are more recent, because of the significantly high number of speciation events (Martín-Bravo et al. 2010). Médail and Diadema (2009) stated that 36% of the Mediterranean refugia were located in mountain ranges. These mountains represent also, in many cases, the southern limit or rear edge of the ranges of numerous central European species (Hampe and Petit 2005) and tree populations located at the edge

of their potential geographical distribution may respond strongly to climate change relatively to those at the core of the range (Carrer et al. 2010). Currently, Mediterranean mountains constitute wet-cool islands of biodiversity in a warm–dry region (Ruiz- Labourdette et al. 2012). Despite the harsh environmental conditions, human presence in Mediterranean mountains has a long history; the use of natural resources through extensive agrosilvopastoral activities has been the main force, which shaped the landscape composition, structure, and functioning (De Aranzabal et al. 2008; Van Eetvelde and Antrop 2004; Vicente-Serrano et al. 2004) creating “cultural landscapes” (Antrop 2005; Naveh and Lieberman 1994). The increasing human influence on Mediterranean ecosystems drove expansion of evergreen sclerophyllous trees in the Mediterranean basin (Vernet 1997; Pons and Quézel 1998; Willcox 1999) and conifers in cold temperate forests (Björkman and Bradshaw 1996).

It has been widely demonstrated that climate and land use changes represent the main drivers affecting mountain ecosystems, particularly at high elevation (Körner and Paulsen 2004; Resco de Dios et al. 2007; Gehrig-Fasel et al. 2007; Chauchard et al. 2007, 2010; Améztegui et al 2010; Ruiz-Labourdette et al. 2012). Changes in temperature and precipitation regimes, and the frequency of extreme events, will have severe consequences on the physical character and on the biological and human communities of mountain areas. Glaciers, snow cover, water storage and flow, are unique features of mountainous areas; however, any changes affecting them will in turn impact on lowland areas (Vogiatzakis 2012). Several regions are potentially more sensitive than others, like the Mediterranean Basin, identified as one of the areas that is most vulnerable to the predicted changes on the basis of a Regional Climate Change Index (RCCI), a comparative index developed based on regional mean precipitation change, mean surface air temperature change, and change in precipitation and temperature interannual variability. The RCCI is designed to identify the most responsive regions to climate change, or Hot-Spots (Giorgi 2006). Many European mountain areas have recorded a significant increase in temperatures since the 1940s (Diaz and Bradley 1997). Forest encroachment in these systems has often been attributed to global warming, which may favour conditions for tree recruitment and growth near or beyond the tree line (Camarero et al. 2006; Batllori and Gutiérrez 2008), and to the variation in precipitation pattern and seasonality. These latter climatic elements are difficult to forecast, although many models predict that summer droughts will become more prevalent (Luterbacher et al. 2006; Peñuelas et al. 2007; IPCC 2007). Where areas above the tree line are limited in size, owing to relatively low summit altitudes, endemic-species hotspots might be affected disproportionately by a rising tree line (Dirnböck et al. 2011; Engler et al. 2011). In a recent study, Ruiz-Labourdette et al. (2012) concluded that large changes in species ranges and forest communities might occur, not only at high elevations within Mediterranean mountains, but also along the entire elevation gradient, throughout this region.

Therefore, Mediterranean mountains might lose their key role as refugia for cold-adapted species. The upward shift of forest tree species, as a function of climate scenarios, could be accompanied by a massive reduction of plant diversity and high community turnover (Thomas et al. 2004; Thuiller et al. 2005; Lenoir et al. 2008; Randin et al. 2009) or a modification of tree-climate correlation (Carrer et al. 2007, 2010). A lengthening of the growing season at mid- to high latitudes (Way 2011; Menzel et al. 2006) and a variation of plant phenology (Cleland et al. 2007) represent other possible responses to recent climate change. All these changes could affect the shifts in ecosystem productivity, with implications for global carbon cycling.

Nevertheless, over the last century European mountain systems have suffered not only global warming but also major demographic, economic and organizational changes (Dirnböck et al. 2003). Therefore, land-use changes must be considered as a major potential factor driving forest expansion, especially in areas exposed to significant human influence, such as the Mediterranean Mountains (Sala et al. 2001; Chauchard et al. 2007; Gehrig-Fasel et al. 2007). Beniston (2003) suggested that, while the causal mechanisms of environmental and climatic change are numerous and complex, the increasing stress imposed by human interference on the natural environment is strictly related to two main factors: economic growth and population growth. The economic level of a country determines to a large extent its resource requirements, in particular energy, industrial commodities, agricultural products and fresh water supply. Rising population levels, on the other hand, can weigh heavily upon the resources available per capita, particularly in less affluent countries. In their quest for timber, fuel and minerals, humans have left their irreversible mark on mountain ecosystems. The current organisation and composition of the forest landscape in the mountains of Europe are strongly determined by legacies of past land-use and management (Koerner et al. 1997; Flinn and Marks 2007). Since the middle of the 19th century, however, these mountain areas have been abandoned, as a result of a rural exodus (Chauchard et al. 2007), and changes in land use encompassed the main processes influencing land-cover variation, ecosystem dynamics and transformation of landscape patterns. In the Mediterranean basin, during the twentieth century, these changes have been characterized by abandonment of marginal areas, grazing cessation and a switch to other materials or energy sources instead of traditional forest resources (Barbero et al. 1990; Debussche et al. 1999). Such land abandonment has led to the encroachment of forest into open areas, or increase in canopy cover of pre-existing forests. The decrease of many semi-natural open habitats in Mediterranean mountains, which had previously been maintained by traditional practices have had a negative impact on the spatial distribution of rare or endemic species (Kiss et al. 2004).

1.2 Focus on the Apennine chain

Climate change in Italian regions, based on historical series (1865-2003), is in accordance with current projections for the Mediterranean Basin (Brunetti et al. 2006; IPCC 2007). On the other hand, past land uses adopted in the Italian mountain ecosystems and the recent abandonment of these traditional practices shaped the landscape structure. In the past centuries, an agro-silvo-pastoral economy affected the landscape of the central-southern Italy, in particular through the transhumance practice, which determined a drastic reduction of shrublands or forests. The human activity affected particularly the transitional belt above the timberline, modifying its original distribution mainly through fire and grazing of natural vegetation, and charcoal production. The abandonment of marginal areas, generally situated in the mountain ecosystems, and grazing cessation allowed a deep expansion and recolonization of shrublands and forests.

Dipped in the heart of the Mediterranean basin, due to their geographical position or their moderate altitude, the Apennines chain represents an ideal site for assessing the effects of global changes on mountain ecosystems. Although this chain stretches from North (Liguria) to South (Calabria) of Italy, it is only in the central Apennines that there are high mountains enough to allow the development of an alpine bioclimatic belt (Blasi et al. 2005). Even though the position of the central Apennines, stretching out in the Mediterranean basin, and xeric landscapes which characterize these mountains, results of the millenary human land use, have led to consider these mountain enclosed within the Mediterranean region, almost the whole of the central Apennines belong to the Temperate Region (Blasi et al. 2003). The relative low altitudes of central Apennines compared to those of the Alps or the Pyrenees chains, together with the strong influence of the Mediterranean climate, means that the alpine belt develops only above an average altitude of 2,400 – 2,500m a.s.l (Baldoni et al. 1999; Blasi et al. 2003). The presence of very high peaks in this chain, apart from allowing the development of a peculiar flora, has played an important role in the preservation of species (and, in some cases, of whole plant community types) with a typical endemic or circumboreal/Arctic-alpine feature (Blasi et al. 2003). Particular is the situation in the Majella massif, where the typical rounded shape of peaks has led to the development of large plateaux even at altitudes exceeding 2.600 m a.s.l., forming a distinct environment that is unique in the Apennine landscape. The Majella massif became part of National Parks world heritage, after decades of strong fights (the first Natural Reserves were created in the 80s), thanks to the Law of 6 December 1991, n. 394 and to the Italian Republic President's Decree of 5 June 1995, constitutive of the Majella National Park (MNP) Authority. This wide and calcareous massif with Morrone, Porrara and Pizzi Mountains, the Valleys and the karst plateaux represent a National Park that, since its geographic position - completely dipped into the

Mediterranean Sea - roughness, size, harshness, its climatic changeableness and the imposing mountains, is surely unique (http://www.parcomajella.it/LgENG/uomo_territorio.asp). During the quaternary glaciations, the higher portion of the Majella massif was covered by an extended ice cap (about 30 km²), which in some places (e.g., the Femmina Morta valley) was up to 200 m thick (Giraudi 1998; Catonica and Manzi 2002). For this reason, the Majella plateaux exhibit some peculiar floristic and coenological characteristics, which suggest a different background to the quaternary development of their vegetation (Blasi et al. 2005). Among these, the krummholz mat [sensu Holtmeier (1981)] composed by mountain pine (*Pinus mugo* Turra ssp. *mugo*), which dominates the treeline ecotone.

1.3 Overall aim of this study

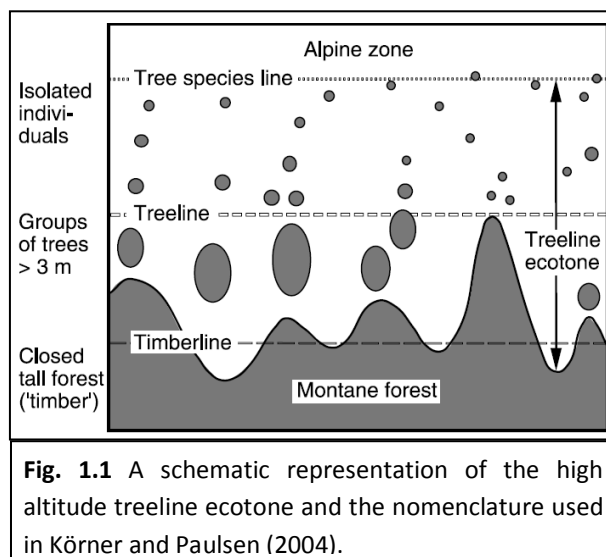
1.3.1 Basic knowledge

Two elements represent the key words of this study:

➤ Treeline ecotone

Körner and Paulsen (2004) suggested a useful description of the treeline ecotone. They stated: “The high altitude limit of forests, commonly referred to as treeline, timberline or forest line (Fig. 1.1) represents one of the most obvious vegetation boundaries. In reality the transition from uppermost closed mountain forests to the treeless alpine vegetation is commonly not a line, but a steep gradient of increasing stand fragmentation, often called the treeline ecotone or the

treeline park land”. Holtmeier, in his recent book, *Mountain Timberlines* (2009), analyzed the great variability of the timberline ecotone which, for the author, represents the same belt occupied by the treeline ecotone in the model of Körner and Paulsen (2004). He wrote: “The existence of these ecotones cannot be primarily and everywhere attributed to human interference and/or unfavourable pedological conditions, but must be explained as the result of the complex influences of the actual and previous climate, fire, biotic factors and site history on tree growth and ecological conditions. In



many cases the existence of a timberline ecotone is the result of oscillations of the climate, persistence of tall (mature) trees and regeneration under changing conditions. A general warming may be followed by advance of the forest to greater altitude and northern latitude, while cooling will cause decay and retreat of the forest in the long-term, followed by change of the site conditions in the former forested area... Moreover, due to the local and regional history of climate, vegetation and, not least, human impact, tree stands at timberline may be different as to successional stage, age classes, composition and ecological dynamics...

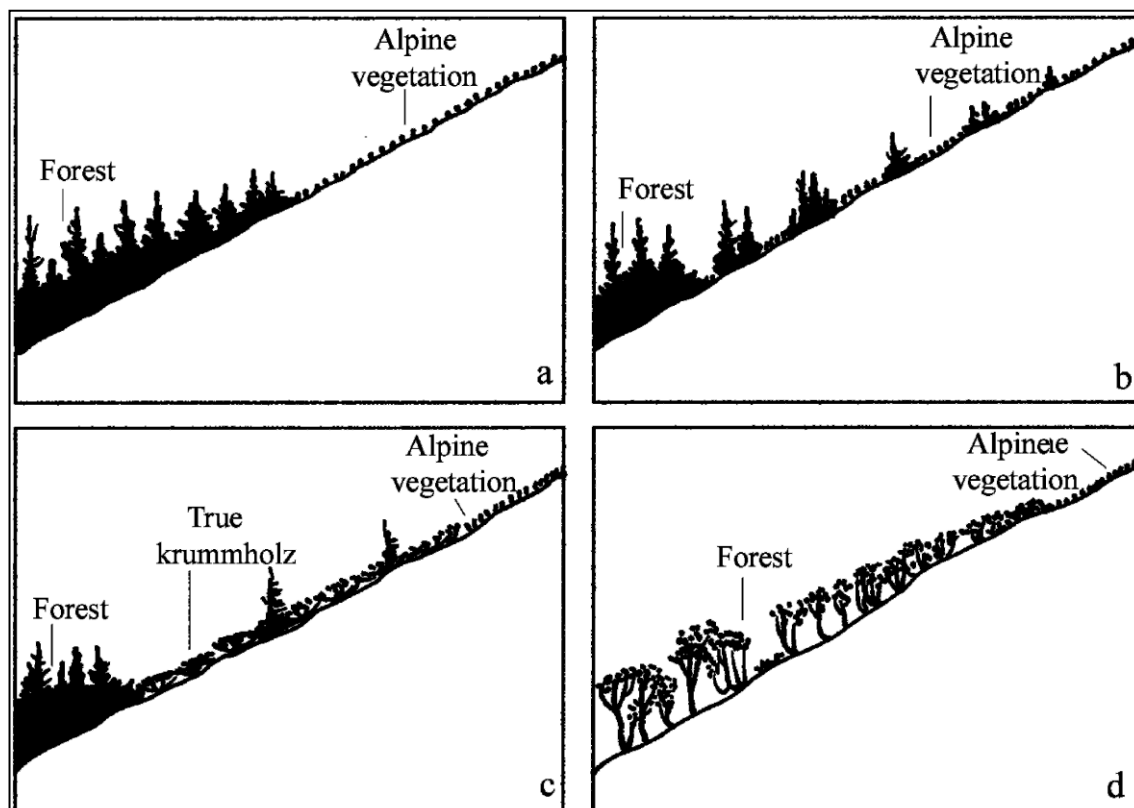


Fig. 1.2 Main types of timberline. a – abrupt forest limit bordering alpine vegetation, b – transition zone (ecotone), c – true krummholz belt (e.g., *Pinus mugo*, *Pinus pumila*) above the upright growing forest, d – gradual transition from high-stemmed forest to crippled trees of the same species bordering alpine vegetation (e.g., *Nothofagus solandri* var. *cliffortioides*). Holtmeier (2009). Modified from Norton and Schönenberger (1984).

Timberline may occur as a line, or as a more or less wide ecotone. Different types (Fig.1.2) may occur in close proximity to each other, even on a single mountainside... Besides climate many other factors such as steep rocky trough walls, talus cones, slope debris and avalanche chutes may prevent the forest from reaching its upper climatic limit. This is the orographic timberline, as is also true for the man-caused (anthropogenic) and edaphic timberlines, are always located more or less far below the elevation to which the forest would advance at the given climatic conditions. Man has influenced mountain forests and timberline in many ways, such as forest pasture, clearing high-elevation forests to create alpine pastures, mining, burning, charcoal production, salt works, timber harvesting for construction wood, fuel and others.”

The treeline ecotone is a transitional belt and as such is very sensitive to minor changes of both environmental and anthropogenic origin. Timberlines –the upper limit of the close forest- in the central Apennines are mostly formed by European beech (*Fagus sylvatica* L.) and are often the result of ancient forest removal to obtain grasslands used initially for wildlife hunting and later for livestock grazing (Piermattei et al. 2012). The presence of a krummholz belt with mountain pine on the treeline ecotone is peculiar of the Majella massif along the entire Apennine chain.

➤ *Pinus mugo* Turra ssp. *mugo*

Pinus mugo Turra ssp. *mugo* is a prostrate conifer 1-3(-5) m tall, with one or more curved trunks; branches long, base lying on the ground (up to 10 m from base), with more or less ascending or erect major branch ends; rarely a tree. The bark is thin, ash-grey-brown to blackish-grey, and splitting in angular scaly plates on old stems. Shoots are uninodal, glabrous and greyish-black to deep red-brown grooved between the decurrent scale-leaves. Buds ovoid-conic, of 6-9 mm, are red-brown very resinous. The needles are grouped in fascicles of two, bright to dark green, and persist 3 to 9 years, slightly twisted, finely serrulate, 28 to 45 mm long, gradually shorter towards the apex of the branch. These plants are usually monoecious, but rarely can be subdioecious. The male cones are grouped at the bottom of the young twigs, are yellow and disperse the pollen from May to June. The female cones are purplish or purple, turn matte dark brown when ripe in late September to October 15-17 months later (Fig. 1.3).



Fig. 1.3 Specimen of prostrate *Pinus mugo* ssp. *mugo* (a). Male cones on 17 June (b) and 29 June (c) and female cones on 17 July (e). Male and female cones in the same twig on 7 July (d).

Its distribution areal (Fig. 1.4) is very fragmented, encompassing Central and Eastern Europe. In Europe it grows from the central Alps near the Swiss-Austrian border to the Carpathians and towards southeast through Yugoslavia and Romania, with an isolated population in the central Italian Apennines. It grows at altitudes of 1400-2500 m, mostly in the high subalpine region at and above the timberline, but also at lower altitudes in peat bogs and frost hollows, exceptionally as low as 200 m in southeast Germany and south Poland (Jovanovic 1986; Christensen 1987). Highest altitudes are reached in the Pirin Mts. of southwest Bulgaria, where it reaches at least 2700 m (M.P. Frankis field notes and herbarium material, Bulgaria 1998). It occurs in Croatia, where it is protected by law. Cold hardiness limit is between -39.9°C and -34.4°C (Bannister and Neuner 2001). The low shrubby growth with a bowed basal section of stem is an adaptation to deep snow cover and avalanches; then branches are erect again during the spring thaw. Mountain pine protects the soil with a strong potential against the erosion and in favour of water retention, especially on the slopes subject to landslides. It stabilizes the snow cover thus hampering the release of avalanches and provides habitat for many species of flora and fauna. Mountain pine krummholz in Italy was subject to the traditional practices only for producing grazing surface, firewood and charcoal.

Fig. 1.4 Above, the distribution map of *Pinus mugo* ssp. *mugo* (black) and ssp. *uncinata* (grey) from Barbéro et al. in Richardson (1998). Red circle indicates the isolated population developed on the Majella massif. Below, a partial view of Mountain pine krummholz within the MNP, showing the south-west slope of the Majella massif (April 2011).



Mountain pine krummholz on the Majella massif (Fig. 1.4) represents the widest stand in Central Apennines, where the species found the southernmost limit of its distributional range, and where in the past should have been the potential natural vegetation of subalpine belt (Migliaccio 1966; Stanisci 1997). Besides, occupying the upper limit of subalpine belt, mountain pine represents a promising species to monitor climatic changes in peculiar environments, with Mediterranean-type climatic patterns and at high elevations.

1.3.2 Dendrochronological studies

Dendrochronology (from the Greek: "chronology"= time, "dendro" = trees) or tree-ring dating is the method of scientific dating based on the analysis of tree-ring growth patterns. This technique was developed during the first half of the 20th century originally by the astronomer A. E. Douglass (Douglass 1940), the founder of the Laboratory of Tree-Ring Research at the University of Arizona. Dendrochronology is an interdisciplinary science, and its theory and techniques can be applied to many applications. The first important principle that governs this science is the *The Uniformitarian Principle* which states that physical and biological processes that link current environmental processes with current patterns of tree growth must have been in operation in the past. In other words, "the present is the key to the past," originally stated by James Hutton in 1785. However, dendrochronology adds a new "twist" to this principle: "the past is the key to the future." In other words, by knowing environmental conditions that operated in the past (by analyzing such conditions in tree rings), we can better predict and/or manage such environmental conditions in the future. Hence, by knowing what the climate-tree growth relationship is in the 20th century, we can reconstruct climate from tree rings well before weather records were ever kept (<http://web.utk.edu/~grissino/principles.htm>).

The theoretical base of the Dendrochronology is the secondary growth of trees, that results from cell division in the *cambium* and causes the stems and roots to thicken. Since the majority of trees have annual growth increment, which is a proper tree ring; the information related to its formation (and factors that influence it) can be represented by the specific characteristics of each ring: width, density and other visual or analytical parameters that can differ one ring from others (Schweingruber 1988). *The Principle of Limiting Factors*, used in dendrochronology, states that rates of plant processes are constrained by the primary environmental variable(s) that is most limiting. For example, precipitation is often the most limiting factor to plant growth in arid and semiarid areas. In these regions, tree growth cannot proceed faster than that allowed by the amount of precipitation, causing the width of the rings (i.e., the volume of wood produced) to be a function of precipitation. In some locations (for example, in higher latitudes and elevations), temperature is often the most

limiting factor (<http://web.utk.edu/~grissino/principles.htm>). However, ring-counting does not ensure the accurate dating of each individual ring. Various techniques are used to “crossdate” wood samples for assuring accurate dating. Crossdating means matching patterns in ring widths or other ring characteristics (such as ring density patterns) among several tree-ring series allow the identification of the exact year in which each tree ring was formed. Dendrochronology is used in different spheres such as archaeology and history (Cook et al. 1995; Baillie 1982), art and criminology, forestry and ecology. Nowadays the use of dendrochronology is very popular in environmental sciences (Schweingruber 1988), particularly with issues related to the climate change (Cook and Kairiukstis 1990; Wigley et al. 1984; Stahl et al. 1998). The climate of our planet is expected to change significantly within the next century. This expectation is mainly based on model calculations, but is also possible to reconstruct the response of the global climate system to certain disturbances by investigating past global changes. Tree-rings represent an excellent archives of such changes. The analysis of tree-rings not only allows for reconstructing the local temperature, the annual precipitation rate or other regional environmental parameters but also the composition of the atmosphere in the past.

1.3.3 Wood anatomy analysis

Trees, as long-living organisms, record ecologically relevant information in their annual rings and hence represent important natural archives for the study of global changes throughout the last millennium (Esper et al. 2002; Cook et al. 2004; Treydte et al. 2006; Trouet et al. 2009). Variation in wood-anatomical characteristics represents adaptive structural solutions adopted by the tree in order to achieve an optimal balance among the competing needs of support, storage and transport under changing environmental conditions and phylogenetic constraints (Chave et al. 2009). The study of wood anatomical structure provides additional ecological information. Variations in xylem anatomy have already been an important source of information in plant sciences (Larson 1994; Gartner 1995). Studying intraspecific variation of woody plants across climatic zones, or between contrasting sites, supplies information about the linkage between ecology (habitat) and functioning (derived from xylem anatomy) (e.g. Carlquist 1975; Baas 1986; Villar-Salvador et al. 1997; Wheeler et al. 2007). Whereas, the study of wood-anatomical variability along tree-ring can elucidate how individual trees and species respond to changing environmental conditions (Schweingruber 1996, 2006). The ability of a genotype to adjust the phenotype over the life of a tree is a result of short-term to long-term physiological responses to environmental variability and can be used to link environment with xylem structure (Fonti et al. 2010).

As described in Fonti et al. 2010 “Tree-ring anatomy is a methodological approach based on dendrochronology and quantitative wood anatomy to assess cell anatomical characteristics (such as conduit size and density, cell wall thickness and tissue percentage) along series of dated tree-rings and to analyse them through time (at the intra- and / or inter-annual level) in order to characterize the relationships between tree growth and various environmental factors. This approach supplements tree-ring based reconstructions of past environmental conditions with novel understanding about the range and strategies of species’ responses and their chances of success, and thus contributes to the evaluation of the impact of predicted climate change on future vegetation dynamics”.

1.3.4 Objectives

Main objective of this study was to analyse the influence of land-use and climatic changes on mountain pine (*Pinus mugo* Turra spp. *mugo*) ecotone dynamics at its southern range margin on the Majella massif, Central Apennines. To achieve this goal, three different studies were carried out with respective three methodological approaches, which correspond to three different spatial scales. From a survey at the landscape scale, which covered the entire Majella massif, we have moved to a survey at the scale of population, based on small plots, to finally analyze at the cellular level the individual of mountain pine.

- 1) The first study analyzed land-cover change of mountain pine at landscape scale. This investigation was carried out in order to assess the effective influence of human practices on mountain pine distribution in the Majella massif, during the last fifty years. A multitemporal analysis of aerial photographs of the study area allowed investigating chronological evolution of mountain pine populations in the Majella massif. While an accurate survey about the number of sheep grazed within the provinces and municipalities of the MNP, provided information regarding the grazing weight in the study area during the last centuries. This work was useful to understand if the treeline ecotone on the Majella massif was man-caused (anthropogenic).
- 2) The second study investigated the dendrochronological potential of mountain pine and its sensitivity to the climatic parameters. The influence of temperature and precipitation on mountain pine growth was assessed together with their variability due to the climate changes. Because of the high elevation of the treeline ecotone and the presence of a krummholz belt in the Majella massif, compared with the rest of the Apennine chain, the

human practices were, most probably, not the only responsible of the treeline ecotone modification. Correlation Function (CF) and moving CF (MCF) were assessed between three mean chronologies of mountain pine and monthly temperature and precipitation. The three mean chronologies were obtained from the growth analysis of mountain pine at three different altitudes.

- 3) The third study examined the mountain pine growth at cellular level. Timing of xylogenesis and intrinsic and extrinsic factor, which influence the wood formation, were defined with particular interest to the climate influence and the reaction wood formation (compression wood in gymnosperm). Mountain pine is a prostrate conifer, which grows adjacent to the soil, its branches need a large amount of compression wood in order to regain the vertical position (Fig. 1.5). How compression wood and opposite wood develop in mountain pine during the growing season has been the subject of the third study.

Chapters 2, 3 and 4 are written as draft manuscripts for publication in scientific journals. This necessitates reiteration of a certain amount of introductory material.



Fig. 1.5 Leaning branches of mountain pine along a deep slope (a); cross-section of a leaning twig showing eccentric pith and wider growth rings in the compression wood (CW) region (b).

1.4 General description of compression wood (CW)

Several reviews on work focusing on CW have been produced (Low 1964; Westing 1965, 1968), but the most comprehensive was the three-volume treatise by Timell (1986), who was one of the pioneering investigators in the field. In addition, a recent Project FP5 - Quality of Life and Management of Living Resources, QLRT-2000-00177 - provided an exhaustive analysis of CW in conifers, in order to characterise its formation and its relevance to timber quality (Gardiner and Macdonald 2005), while a Ph.D. Thesis stressing the relationship between chemical composition and severity of CW provided a complete description of chemical feature of CW (Nanayakkara 2007). Here a brief description of the principal features, which characterise the CW, including chemical, physical and anatomical properties, is presented based on the works above mentioned.

CW is formed on the underside of leaning trees and branches. Its effect is manifested by a slow change in the orientation of the stem or branch. Formation of CW is generally considered to be a gravitational response brought about by the redistribution of auxins.

The structure of CW was defined in comparison with normal wood (NW): key differences of its anatomical appearance are shorter and rounder tracheids of CW, with distorted tips and thick cell walls. CW tracheids lack of a tertiary cell wall (S3) layer. Most thickening and enhanced lignification of the outer secondary wall, designed as S_{2L} layer by Côté et al. (1968), shape the tracheids with a more rounded appearance in cross section, and then the presence of intracellular spaces (IS), which results from less connection between the tracheids. The inner part of the S2 layer presents a higher microfibril angle (in the range 30-50°) and is deeply fissured so that the wall consists of helical cavity. The cavities follow the direction of the microfibrils (Fig. 1.6).

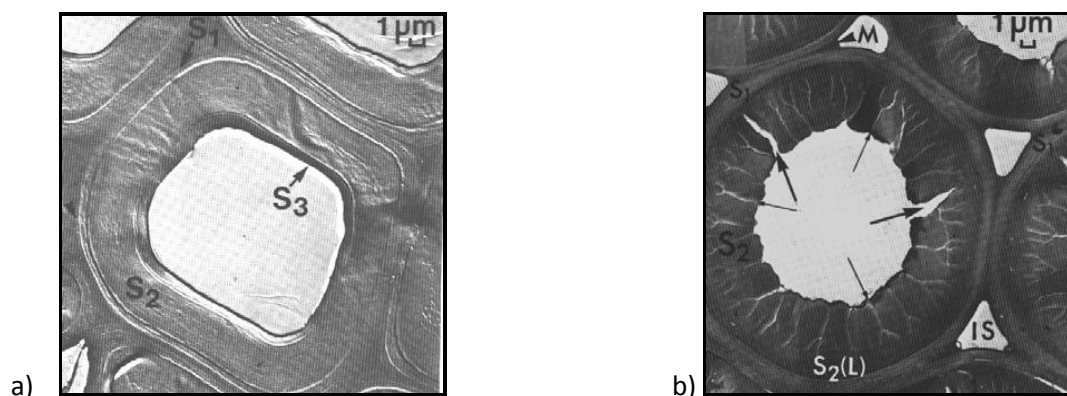


Fig. 1.6 Cross-section of a normal wood tracheid (a), and a compression wood tracheid (b). Middle lamella (M); Primary wall (S1); Secondary wall (S2); Outer secondary wall (S_{2L}); Tertiary wall (S3); Inter cellular spaces (IS); Helical cavities (small arrows); Drying checks (large arrows). Images from Timell (1986) in *Compression Wood in Gymnosperms* Vol. 1.

All degrees of CW may be encountered from moderate forms to extreme types (severe CW). Every sample may not have all of the features described above. In extreme cases, CW may develop all the features. Mild CW formation is common in fast-growing conifer species. It can be found:

- on the lower side of the stems of slightly-leaning trees
- on the lower side of tree stems that have been strongly tilted for a short period
- on one side of strongly-leaning trees
- in the transition zone between normal wood and CW.

Microscopy is the most accurate method for detecting CW. It allows a clear distinction to be made between mild and severe forms (Fig. 4.3). Mild CW formation exhibits more variable anatomical features than severe CW. In mild cases tracheids resemble those of normal wood in their cross-sectional shape with slightly rounded appearance. According to Yumoto et al. (1983), the first feature to appear in CW formation is increased lignification of the S2_L. This occurs first in the corners and later a continuous layer is formed (Fig. 1.7).

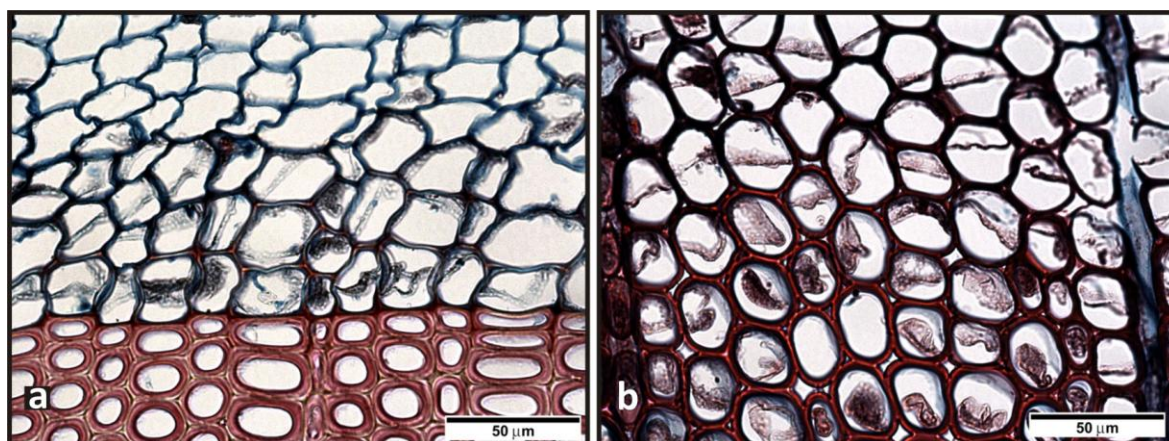


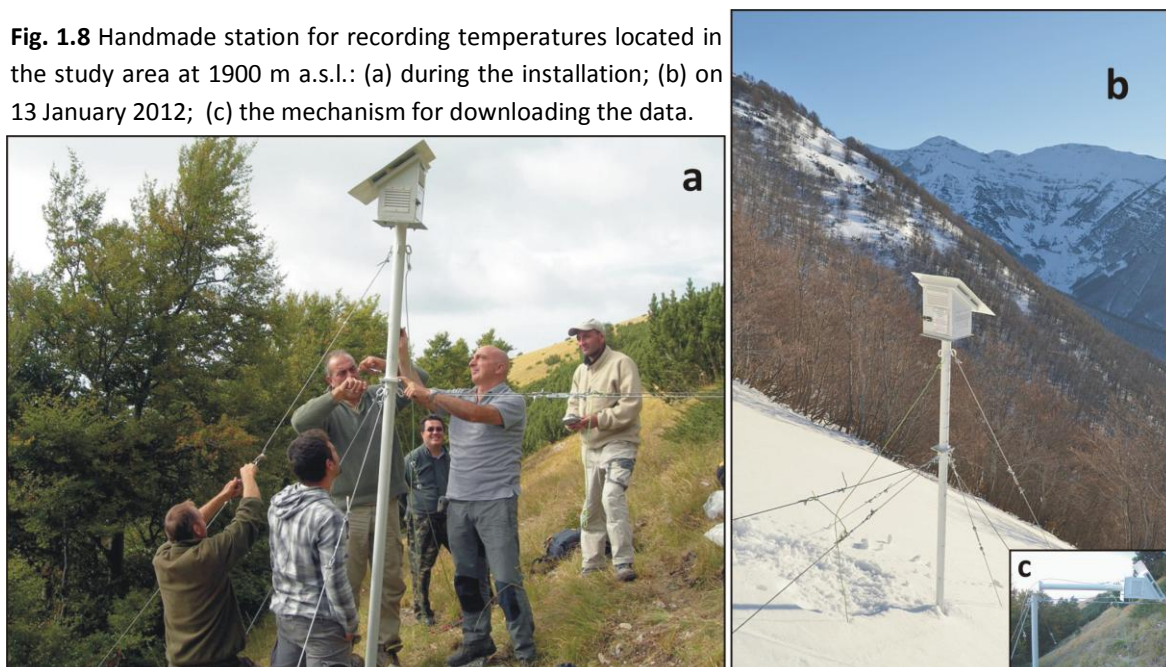
Fig. 1.7 Progressive lignification of tracheids in CW, starting from the corner of the semi-rounded cells [(a) on 1 June] and then continues along the rounded border of cells [(b) on 10 June].

In chemical terms, the same substances are present in both CW and NW but in different amounts. Compression wood contains 30-40% more lignin and 20-25% less cellulose than normal wood but the amount of hemicelluloses is not changed. In the lignin in compression wood the amount of some polysaccharides also differs. The chemical composition and anatomical features of CW influence its physical properties. The difference in the chemical configuration of the lignin does not have a major effect on the mechanical properties of wood, but they can be of importance in the pulp and paper industry.

1.5 Climate data collection

Data-gathering of temperature and precipitation within the study area was crucial to the realization of this study. Climate at the treeline ecotone represents one of the most important limiting factors for the plants growth. However, in many cases, it is also one of the factors more difficult to monitor. The scarcity of meteorological stations located at high altitudes is often the main obstacle for the climate characterization of these areas. Short and incomplete climatic series often prevent the construction of specific climatic trends, and then the monitoring of climate-growth relationships. In this study, an important work on the collection and management of climate data was performed (Section 3.2.3; Annex I). Climate-growth relationships of mountain pine were assessed by gridded climate dataset, downloaded from the KNMI Climate Explorer web site (http://climexp.knmi.nl/selectfield_obs2.cgi?id=someone@somewhere). In particular, temperature and precipitation data used in Chapter 3 belong to the Climatic Research Unit (CRU) time-series datasets of variations in climate with variations in other phenomena. Climate influence on xylogenesis was assessed by using temperature data recorded by a datalogger located in the study area. At the beginning of the growing season a handmade weather station was constructed in collaboration with the Corpo Forestale dello Stato – CFS. This station was installed in a small clearing between the upper limit of beech forest and the lower limit of mountain pine krummholz, at 1900 m a.s.l. (Fig. 1.8). The temperature logger was placed at 2.5 m above the ground, protected from direct solar radiation by a white frame. Data recording is still in progress, in order to monitoring the temperature trend at high elevation. Occasionally is required a maintenance operation for downloading the recorded data and controlling the correct functioning of the instrument.

Fig. 1.8 Handmade station for recording temperatures located in the study area at 1900 m a.s.l.: (a) during the installation; (b) on 13 January 2012; (c) the mechanism for downloading the data.



References

- Améztegui A., Brotons L., Coll L. 2010. Land-use changes as major drivers of mountain pine (*Pinus uncinata* Ram.) expansion in the Pyrenees. *Global Ecology and Biogeography* 19: 632–641.
- Antrop M. 2005. Why landscapes of the past are important for the future. *Landscape and Urban Planning* 70(1-2): 21-34.
- Baas P. 1986. Ecological patterns of xylem anatomy. In *On the economy of plant form and function*, Givnish T.D. (ed.). Cambridge, UK: Cambridge University Press, pp. 327–352.
- Baillie M.G.L. 1982. Tree-Ring Dating and Archaeology. Croom Helm Ltd, London; Great Britain.
- Baldoni M., Biondi E. and Frattaroli R. 1999 Caratterizzazione bioclimatica del Gran Sasso d'Italia. *Braun-Blanquetia* 16: 7-21.
- Bannister P. and Neuner G. 2001. Frost resistance and the distribution of conifers, in *Conifer cold hardiness*, F.J. Bigras and S.J. Colombo (eds.). Dordrecht: Kluwer Academic Publishers, pp. 3-22.
- Barbero M., Bonin G., Loisel R., Quézel P. 1990. Changes and disturbances of forest ecosystems caused by human activities in the western part of the Mediterranean basin. *Vegetation* 87: 151–73.
- Batllo E. and Gutiérrez E. 2008. Regional tree line dynamics in response to global change in the Pyrenees. *Journal of Ecology* 96: 1275–1288.
- Beniston M. 2003. Climatic change in mountain regions: a review of possible impacts. *Climatic Change* 59: 5–31.
- Björkman L. and Bradshaw R.H.W. 1996. The immigration of *Fagus sylvatica* L. and *Picea abies* (L.) Karst. into a natural forest stand in southern Sweden during the last 2000 years. *Journal of Biogeography* 23: 235–244.
- Blasi C., Di Pietro R., Fortini P., Catonica, C. 2003. The main plant community types of the alpine belt of the Apennine chain. *Plant Biosystems* 137(1): 83-110.
- Blasi C., Di Pietro R., Pelino G. 2005. The vegetation of alpine belt karst-tectonic basins in the Central Apennines. *Plant Biosyst* 139: 357-385.
- Brunetti M., Maugeri M., Monti F., Nanni T. 2006. Temperature and precipitation variability in Italy in the last two centuries from homogenised instrumental time series. *International Journal of Climatology* 26: 345–381.
- Camarero J.J., Gutiérrez E., Fortin M.J. 2006 Spatial patterns of plant richness across treeline ecotones in the Pyrenees reveal different locations for richness and tree cover boundaries. *Global Ecology and Biogeography* 15: 182–191.

- Carlquist S 1975. Ecological strategies of xylem evolution. Berkeley, CA, USA: University of California Press.
- Carrer M., Nola P., Eduard J.L., Motta R., Urbinati C. 2007. Regional variability of climate-growth relationships in *Pinus cembra* high elevation forests in the Alps. *Journal of Ecology* 95: 1072–1083.
- Carrer M., Nola P., Motta R., Urbinati C. 2010. Contrasting tree-ring growth to climate responses of *Abies alba* toward the southern limit of its distribution area. *Oikos* 119: 1515–1525.
- Catonica C. and Manzi A. 2002. L'influenza della storia climatica e geologica recente sulla Flora d'alta quota dei gruppi montuosi del Gran Sasso e della Majella (Appennino Centrale). *Rivista Piemontese di Storia Naturale* 23: 19–29.
- Chauchard S., Beilhe F., Denis N., Carcaillet C. 2010. An increase in the upper tree-limit of silver fir (*Abies alba* Mill.) in the Alps since the mid-20th century: A land-use change phenomenon. *Forest Ecology and Management* 259: 1406–1415.
- Chauchard S., Carcaillet C., Guibal F. 2007. Patterns of land-use abandonment control tree-recruitment and forest dynamics in Mediterranean mountains. *Ecosystems* 10: 936–948.
- Chave J, Coomes D, Jansen S, Lewis SL, Swenson NG, Zanne AE. 2009. Towards a worldwide wood economics spectrum. *Ecology Letters* 12: 351–366.
- Christensen K.I. 1987. Taxonomic revision of the *Pinus mugo* complex and *P. × rhaetica* (*P. mugo* × *sylvestris*) (Pinaceae). *Nordic J. Botany* 7: 383–408.
- Cleland E.E., Chuine I., Menzel A., Mooney H. A, Schwartz M.D. 2007. Shifting plant phenology in response to global change. *Trends in ecology and evolution* 22: 357–65.
- Cook E.R. and Kairiuksis L.A. 1990. The Statistics of Ring-Width and Climatic Data, Chapter 6. International Institute for Applied Systems Analysis, the Netherlands.
- Cook E.R., Briffa K.R., Meko D.M., Graybill D.A., Funkhouser G. 1995. The segment length curse in long tree-ring chronology development for paleoclimatic studies. *The Holocene* 5(2): 229–237.
- Cook ER, Woodhouse CA, Eakin CM, Meko DM, Stahle DW. 2004. Long-term aridity changes in the western United States. *Science* 306: 1015–1018.
- Côté W.A., Day A.C., Timell T.E. 1968. Studies on compression wood VII. Distribution of lignin in normal and compression wood of tamarack *Larix laricina* (Du Roi, K. Koch). *Wood Science and Technologies* 2: 13–37.
- De Aranzabal I., Schmitz M.F., Aguilera P., Pineda P. 2008. Modelling of landscape changes derived from the dynamics of socio-ecological systems. A case of study in a semiarid Mediterranean landscape. *Ecological Indicators* 8: 672–685.
- Debussche M., Lepart J., Dervieux A. 1999. Mediterranean landscape changes: evidence from old postcards. *Global Ecology and Biogeography* 8:3–15.

- Diaz H.F. and Bradley R.S. 1997. Temperature variations during the last century at high elevation sites. *Climatic Change* 36: 253–279.
- Dirnböck T., Dullinger S., Grabherr G. 2003. A regional impact assessment of climate and land-use change on alpine vegetation. *Journal of Biogeography* 30: 401–417.
- Dirnböck T., Essl F., Rabitsch W. 2011. Disproportional risk for habitat loss of high-altitude endemic species under climate change. *Global Change Biology* 17: 990–996.
- Douglass A.E. 1940. Tree-Ring Dates from the Foresdale Valley, East-Central Arizona. *Tree-Ring Bulletin* 7(2).
- Engler R., Randin C., Thuiller W. et al. (2011) 21st century climate change threatens mountain flora unequally across Europe. *Global Change Biology* 17: 2330–2341.
- Esper J, Cook ER, Schweingruber FH. 2002. Low-frequency signals in long tree-ring chronologies and the reconstruction of past temperature variability. *Science* 295: 2250–2253.
- Flinn K.M., Marks P.L. 2007. Agricultural legacies in forest environments: tree communities, soil properties, and light availability. *Ecological Applications* 17: 452–463.
- Fonti P., Von Arx G., García-González I., Eilmann B., Sass-Klaassen U., Gärtner H., Eckstein D. 2010. Studying global change through investigation of the plastic responses of xylem anatomy in tree rings. *The New phytologist* 185, 42–53.
- Gardiner B. and Macdonald E. 2005. Compression wood in conifers-the characterisation of its formation and its relevance to timber quality, in European Union - Framework Programme FP5 - Quality of Life and Management of Living Resources, QLRT-2000-00177, pp 376.
- Gartner BL. 1995. Plant stems: physiological and functional morphology. New York, NY, USA: Academic Press.
- Gehrig-Fasel J., Guisan A., Zimmermann N.E. 2007. Tree line shifts in the Swiss Alps: Climate change or land abandonment? *Journal of Vegetation Science* 18: 571–582.
- Giorgi F. 2006. Climate change hot-spots. *Geophysical Research Letters* 33: L08707.
- Giraudi C. 1998. Nuovi dati sul glacialismo della Majella (Abruzzo, Italia centrale). *Italian Journal* 11(2): 265–271.
- Hampe A. and Petit J. 2005. Conserving biodiversity under climate change, the rear edge matters. *Ecology Letters* 8: 461–467.
- Holtmeier F.K. 1981. What does the term 'krummholz' really mean? Observations with special reference to the Alps and the Colorado Front Range. *Mountain Research and Development* 1: 253–260.
- Holtmeier F.K. 2009. Mountain Timberlines: Ecology, Patchiness, and Dynamics Advances, in Global Change Research, vol. 36. Second edition, *Springer Science + Business Media B.V.*, pp.438.

- IPCC 2007. Climate change 2007: impacts, adaptation and vulnerability. Southern Europe. Working Group II *Contribution to the Intergovernmental Panel on Climate Change, United Nations Environmental Program*, Cambridge University Press, Cambridge.
- Jovanovic B. 1986. "*Pinus mugo*" in Flora Srbije. *Belgrade: Serbian Academy of Sciences and Arts*.
- Kiss L., Magnin F., Torre F. 2004. The role of landscape history and persistent biogeographical patterns in shaping the responses of Mediterranean land snail communities to recent fire disturbances. *Journal of Biogeography* 31:145–57.
- Koerner W., Dupouey J.L., Dambrine E., Benoît M. 1997. Influence of past land use on the vegetation and soils of present day forest in the Vosges mountains, France. *Journal of Ecology* 85: 351–358.
- Körner C. and Paulsen J. 2004. A world-wide study of high altitude treeline temperatures. *Journal of Biogeography* 31: 713–732.
- Larson PR. 1994. The vascular cambium: development and structure. Berlin Heidelberg New York: Springer Verlag.
- Lenoir J., Gégout J.C., Marquet P.A., De Ruffray P., Brisse H. 2008. A significant upward shift in plant species optimum elevation during the 20th century. *Science* 320: 1768–1771.
- Lionello P., Malanotte-Rizzoli P., Boscolo R., Alpert P., Artale V., Li L., Luterbacher J., May W., Trigo R.M., Tsimplis M., Ulbrich U., Xoplaki. E. 2006. The Mediterranean climate, an overview of the main characteristics and issues, in *Mediterranean climate variability*, P. Lionello, P. Malanotte-Rizzoli and R. Boscolo (eds.). Elsevier, Amsterdam, pp. 1–26.
- Liutsko L. 2008. What trees tell us. Dendrochronological and statistical analysis of the data. Department of Statistics, Biology Faculty, University of Barcelona [Thesis].
- Low A. J. 1964. Compression wood in conifers. A review of literature. *Forestry Abstracts* 25(3):33.
- Luterbacher J. et al. 2006. Mediterranean climate variability over the last centuries: A review, in *The Mediterranean Climate: an overview of the main characteristics and issues*, P. Lionello, P. Malanotte-Rizzoli and R. Boscolo (eds.). Elsevier, Amsterdam, pp. 27–148.
- Marchetti M., Tognetti R., Lombardi F., Chiavetta U., Palumbo G., Sellitto M., Colombo C., Iovieno P., Alfani A., Baldantoni D., Barbati A., Ferrari B., Bonacquisti S., Capotorti G., Copiz R., Blasi C. 2010. Ecological portrayal of old-growth forests and persistent woodlands in the Cilento and Vallo di Diano National Park (southern Italy). *Plant Biosystems* 144: 130–147.
- Martín-Bravo S., Valcárcel V., Vargas P., Luceño M. 2010. Geographical speciation related to Pleistocene range shifts in the western Mediterranean mountains. *Taxon* 59: 466–482.
- Médail F. and Diadema K. 2009. Glacial refugia influence plant diversity patterns in the Mediterranean Basin. *Journal of Biogeography* 36: 1333–1405.

- Médail F. and Quézel P. 1999. Biodiversity Hotspots in the Mediterranean Basin: Setting Global Conservation Priorities. *Conservation Biology* 13 (6):1510-1513.
- Menzel A., Sparks T.H., Estrella N. et al. 2006. European phenological response to climate change matches the warming pattern. *Global Change Biology* 12: 1969–1976.
- Metzger M.J., Bunce R.G.H., Jongman R.H.G., Múcher C.A., Watkins J.W. 2005. A climatic stratification of the environment of Europe. *Global Ecology and Biogeography* 14: 549–563.
- Migliaccio F. 1966. La vegetazione a *Pinus pumilio* della Majella. *Annali di Botanica* (Roma) 28: 539–550.
- Myers N, Mittermeier R.A., Mittermeier C.G., Da Fonseca G.A.B., Kent J. 2000. Biodiversity hotspots for conservation priorities. *Nature* 403: 853–858.
- Nanayakkara B. 2007. Chemical characterisation of Compression Wood in plantation grown *Pinus radiata*. Ph.D. dissertation at the University of Waikato, New Zealand.
- Naveh Z. and Lieberman A. 1994. Landscape ecology, theory and application. New York: Springer-Verlag.
- Peñuelas J., Prieto P., Beier C., Cesaraccio C., De Angelis P., De Dato G., Emmett B., Estiarte M., Garadnai J., Gorissen A., Lang E., Kröel-Dulay G., Llorens L., Pellizzaro G., Riis-Nielsen T., Schmidt I., Sirca C., Sowerby A., Spano D., Tietema A. 2007. Response of plant species richness and primary productivity in shrublands along a north-south gradient in Europe to seven years of experimental warming and drought: reductions in primary productivity in the heat and drought year of 2003. *Global Change Biology* 13:2563-2581.
- Piermattei A., Renzaglia F., Urbinati C. 2012. Recent expansion of *Pinus nigra* Arn. above the timberline in the central Apennines, Italy. *Annals of Forest Science* 69, 509–517.
- Pons A., Quézel P. 1998. A propos de la mise en place du climat méditerranéen. *Comptes Rendus de l'Académie des Sciences Paris, Série Sciences de la Terre et des Planètes* 327:755–760.
- Price L.W. 1981. Mountains and Man: a Study of Process and Environment. University of California Press.
- Randin C.F., Engler R., Normand S. et al. 2009. Climate change and plant distribution: local models predict high-elevation persistence. *Global Change Biology* 15: 1557– 1569.
- Regato P. and Salman R. 2008. Mediterranean Mountains in a Changing World. *Guidelines for Developing Action Plans. IUCN*.
- Resco de Dios V., Fischer C., Colinas C. 2007. Climate Change Effects on Mediterranean Forests and Preventive Measures. *New Forests* 33: 29–40.
- Ruiz-Labourdette D., Nogués-Bravo D., Ollero H.S., Schmitz M.F., Pineda F.D. 2012. Forest composition in Mediterranean mountains is projected to shift along the entire elevational gradient under climate change. *Journal of Biogeography* 39: 162–176.

- Sala O.E., Chapin III F.S., Huber-Sannwald E. 2001. Potential biodiversity change: global patterns and biome comparisons, in *Global biodiversity in a changing environment: scenarios for the 21st century*, F.S. Chapin III, O.E. Sala and E. Huber-Sannwald (eds.). Springer-Verlag, New York, pp. 351–368.
- Schweingruber F.H. 1988. Tree rings: bases and applications of dendrochronology. D. Reidel publishing company; Dordrecht, Holland.
- Schweingruber F.H. 1996. Tree rings and environment – dendroecology. Bern, Switzerland: Paul Haupt.
- Schweingruber F.H. 2006. Wood structure and environment. Berlin Heidelberg, New York, NY, USA: Springer-Verlag.
- Stahle D.W. et al. 1998. Experimental Dendroclimatic Reconstruction of the Southern Oscillation. *Bulletin of the American Meteorological Society* 79(10): 2137-2152.
- Stanisci A. 1997. Gli arbusteti altomontani dell'Appennino centrale e meridionale. *Fitosociologia* 34: 3-46.
- Thomas C.D., Cameron A., Green R.E. et al. 2004. Extinction risk from climate change. *Nature* 427: 145–148.
- Thuiller W., Lavorel S., Araujo M.B., Sykes M.T., Prentice I.C. 2005. Climate change threats to plant diversity in Europe. *Proceedings of the National Academy of Science* 102: 8245– 8250.
- Timell T. E. 1986. Compression Wood in Gymnosperms, Volume 1-3, Springer-Verlag, Berlin.
- Treydte K, Schleser GH, Helle G, Frank DC, Winiger M, Haug GH, Esper J. 2006. The twentieth century was the wettest period in northern Pakistan over the past millennium. *Nature* 440: 1179–1182.
- Trouet V, Esper J, Graham NE, Baker A, Scourse JD, Frank DC. 2009. Persistent positive North Atlantic Oscillation mode dominated the Medieval Climate Anomaly. *Science* 324: 78–80.
- Van Eetvelde V. and Antrop M. 2004. Analyzing structural and functional changes of traditional landscapes—two examples from Southern France. *Landscape and Urban Planning* 67(1–4): 79–95.
- Vernet J.L. 1997. L'homme et la forêt méditerranéenne de la préhistoire à nos jours. Editions Errance, Paris, France.
- Vicente-Serrano S.M., Lasanta T., Romo A. 2004. Analysis of spatial and temporal evolution of vegetation cover in the Spanish central Pyrenees: Role of human management. *Environmental Management* 34(6): 802–818.
- Villar-Salvador P, Castro-Díez P, Pérez-Rontomé C, Montserrat-Martí G. 1997. Stem xylem features in three Quercus (Fagaceae) species along a climatic gradient in NE Spain. *Trees – Structure and Function* 12: 90–96.

- Vogiatzakis. I.N. 2012. Mediterranean Mountain Environments. First Edition, John Wiley & Sons, Ltd., pp. 216.
- Way D.A. 2011. Tree phenology responses to warming: spring forward, fall back? *Tree physiology* 31: 469–71.
- Westing A.H. 1965. Formation and function of compression wood in gymnosperms. *The Botanical Review* 31: 381.
- Wheeler EA, Baas P, Rodgers S. 2007. Variations in dicot anatomy: a global analysis based on the Insidewood database. *IAWA Journal* 28: 229– 258.
- Wigley T.M.L., Briffa K.R., Jones P.D. 1984. On the Average Value of Correlated Time Series, with Applications in Dendroclimatology and Hydrometeorology. *Journal of Climate and Applied Meteorology* 23: 201-213.
- Willcox G. 1999. Charcoal analysis and Holocene vegetation history in southern Syria. *Quaternary Science Reviews* 18:711–716.
- Yumoto M., Ishida S., Kukazawa K. 1983. Studies on the formation and structure of the compression wood cells induced by artificial inclination in young trees of *Picea glauca*. IV. Gradation of severity of compression wood tracheids. *Res Bull Coll Exp For, Hokkaido Univ* 40:409–454.

SECOND CHAPTER:

Is land abandonment affecting forest dynamics at high elevation in Mediterranean mountains more than climate change?

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Article in press by *Plant Biosystems*



ABSTRACT

Global change is leaving a fingerprint on the appearance, structure, and productivity of the tree-line ecotone, modifying patterns of mountain ecosystems. In order to implement correct policies for managing natural resources, we examine how climate change interrelated with land-use abandonment could shape mountain forests at their upper limit in a Mediterranean environment and how patterns of tree growth and periods of tree establishment guide the interpretation of global change effects on tree-line dynamics. We reconstructed the population dynamics of mountain pine (*Pinus mugo* Turra spp. *mugo*) in the subalpine belt of the Majella National Park (Italy). In a test area of 14440 ha, proposed as a pilot study site for long-term ecological monitoring, temporal and spatial mountain pine distribution were examined since 1954 by historical aerial ortophotos. Multitemporal maps documented the expansion upward (1 m per year) and downward (3 m per year) of mountain pine. Mountain pine started to expand upwards into the formerly tree-free grassland in early 1900s, in association with a decline of the local human population and livestock. Land-use change was the major driving force of vegetation dynamics at the tree line in the Majella massif.

2.1 Foreword

One of the biggest gains of the global conference Rio+20 on sustainable development has been the pledge to protect mountain ecosystems. The Rio+20 outcome document encourages states and organizations to take concrete actions since mountains offer solutions both to kick-start a sustainable and equitable Green Economy and demonstrate inclusive development models that have reduced poverty and promoted social and gender equity in both upstream and downstream contexts (Kohler et al. 2012). The benefits deriving from mountain regions are essential for sustainable development of countries facing the Mediterranean Basin; in fact mountain ecosystems play a crucial role in providing water resources to a large portion of the population. Mountain ecosystems are, however, fragile and particularly vulnerable to the adverse impacts of climate change, deforestation and forest degradation, land use change, land degradation, and natural disasters. Local communities have developed sustainable uses of mountain resources, but they are often marginalised. Pisanelli et al (2012) in a recent study conducted in a mountain area of central Italy stress the awareness of social, cultural and environmental constraints/potentialities of the local communities. They also highlight that continued efforts would be required especially to satisfy expectation of young people concerning the territorial development. The research community is invited to boost networking with effective involvement and sharing of experience of all relevant stakeholders, by strengthening existing cooperation, as well as exploring new development strategies.

In Southern Europe, the legacy of Mediterranean civilizations have led, mainly through fire and grazing of natural vegetation, to the overexploitation and transformation of pristine ecosystems into the present human-made landscapes. The landscape in these areas was artificially modified by man for the purpose of enhancing agriculture, forestry and other productive activities. Such traditional techniques and practices represent a relevant value of this cultural landscape (Agnoletti 2007). Industrialization processes of Europe during the 19th and 20th centuries have triggered deep socioeconomic shifts, including rural exodus and declines of traditional practices based on small-scale agriculture, pastoralism and forest resource utilization (Blondel and Aronson 1999). These changes in land-use were characterized by the abandonment of marginal areas, generally situated in the mountains, where land-cover change, grazing cessation, and switch from habitual forest resources to other construction material or fuel type have transformed the landscape patterns deeply (Boden et al. 2010; Debussche et al. 1999).

The principal effect in the Italian countryside has been the spontaneous return of woodlands on land previously used for agriculture and pasture. This patchy process is influenced by the presence of former agricultural structures, such as terraces or walls, and crops, including fruit trees. This secondary succession is currently accelerating in many areas of the Italian Peninsula, acutely in mountainous areas (Motta et al. 2006; Boden et al. 2010). The same is happening in the central Apennines, where 2400 m a.s.l. represent the lower border of the subalpine/alpine ecocline (Blasi et al. 2003). In this natural test site, the typical tree line with a compact frame of beech forest is certainly anthropogenic, as the result of centuries of human activities (grazing, burning and logging). The Majella massif, in particular, has a wide altitudinal range available for tree recruitment in the alpine belt; it is the only area in the Apennines with large patches above 2000 m a.s.l., consisting of gentle slopes and large plateaus (Blasi et al. 2005). The presence of woodland patches [krummholz, sensu Holtmeier (1981)] dominated by mountain pine (*Pinus mugo* Turra spp. *mugo*) at the tree line on dry plateaus of the Majella massif resembles the subalpine-alpine ecotone on the Alps, and suggests that in the past a real climatically induced tree line must have been occurred somewhere sporadically on the Apennines.

Forest expansion in the Majella massif can be hypothetically attributed to three major (interrelated) issues: (i) cessation of grazing, burning and logging (especially after the foundation of the Majella National Park in the 1995), which influence present land use; (ii) increase in summer air temperatures, which affects tree growth at the natural boundary; (iii) changes in natural disturbances, which create opportunities for the establishment of new individuals. Li Dai (2010) predicted krummholz expansion on the Majella massif using a model with neighbourhood and

environmental variables. Palombo (2013) reconstructed the population dynamics of mountain pine back in time by dating periods of past tree establishment, to understand tree-establishment patterns over the past 100 years, and found that tree growth of mountain pine was positively influenced by current spring temperature and summer precipitation. We aimed at investigating the population dynamics of mountain pine in the Majella National Park through a multitemporal comparison, to interpret recent tree-line changes due to changes in land use. To achieve this goal we examined: (i) spatial patterns of tree establishment in current tree-line forests; (ii) evidence for recent establishment above the tree line; and (iii) socio-economic changes in the alpine belt.

2.2 A pilot site for long-term monitoring of the tree line on the Apennines

The Majella massif (Abruzzo Region, central Italy - Fig. 2.1), reaching 2793 m a.s.l. with Mt. Amaro, creates a ridge with a latitudinal span between 42°09'33" N and 42°00'14" N, 32 km from the Adriatic Sea. The Majella massif is composed almost exclusively by large layers of limestone, with the highest part characterised by a wide summit surface weakly sloped, of structural origin, bordered by steep inclinations, incised by deep valleys. Karstic phenomena feature in depth the whole massif. Secondary montane grasslands, currently unutilized, prevail on south-eastern to south-western slopes. On satellite imagery (e.g. Arc Globe at 0.5 km resolution), the Majella altipiano appears as the largest ($\approx 100 \text{ km}^2$) barrens in the Apennines. On the ground, these alpine barrens include patches of grassland and open low-vegetation, often featuring tussock and cushion growth forms (Blasi et al. 2005, van Gils et al. 2012).

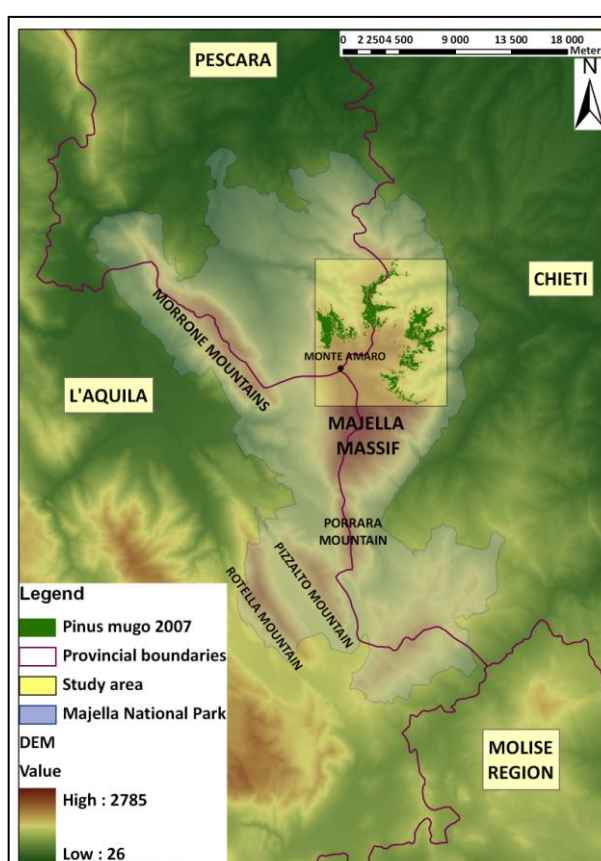


Fig. 2.1 Location of the MNP (blue-grey) in the Abruzzo Region. In yellow the study area and in dark green the mountain pine distribution, in 2007.

The mountain pine is rare in the Apennines, and is considered the potential vegetation of the subalpine belt in central Apennines (Migliaccio 1966). The largest stands are in the Majella massif, above the beech forest limit, where mountain pine finds its ecological optimum in the north facing

altitudinal range. The subalpine/alpine belt of Majella massif was subjected to the traditional summer pasturing until the 1950s and, since high-altitude pasture abandonment, upward moving of mountain pine dwarf-shrubland up to 2450 m a.s.l. has occurred. Mountain pine is a drought-tolerant species that grows successfully on carbonate-rich soils, and current climatic changes might thus be favourable to its expansion. Having slopes with prairies up to an elevation of 2400 m a.s.l., the Majella massif is probably the most suited Apennines' site for investigation on the effects of global warming and land-use change in Mediterranean mountains.

The highest long-term meteorological station of central Apennines is Campo Imperatore (2125 m a.s.l.) with 3.6 °C as mean annual temperature and 1613 mm as total annual precipitation (1960–1990). Stanisci et al. (2005), analysing the changes in species richness along an altitude gradient (2405–2730 m a.s.l.), found that 70% of species does not reach the highest summit and only 11% of the overall flora is shared by all summits of the massif; a drop in mean temperature was observed at soil level, along the same gradient from 3.1 to about 0 °C. The study area is 14440 ha wide and corresponds to the subalpine-alpine humid type with mountain pine woodlands as potential natural vegetation of slopes [habitat 4070 * bushes of *Pinus mugo* and *Rhododendron hirsutum* (Mugo-Rhododendretum hirsuti)], referring to the alliance Epipactido atropurpureae-Pinion mugo (Stanisci 1997).

2.3 Spatial patterns of tree-line mountain pine forests

The analysis of recent and historical aerial-photo coverage of the area enabled us to investigate the dynamics of mountain pine populations in the Majella massif (Table 2.1). Two digital co-registered colour orthophotos were available: one of summer 1999, with a spatial resolution of 1 m, and one of summer 2007, with a spatial resolution of 0.5 m.

Table 2.1. Technical details of the aerial photographs used in the analysis and achieved results.

Year	1954	1991	1999	2007
Scale	1:33000	1:33000	Pixel 1 m	Pixel 0.5 m
Format	Print 23x23 cm black/white	Print 23x23 cm black/white	Digital, real colours	Digital, real colours
Resulting mountain pine area	693 ha	1196 ha	1362 ha	1423 ha
Minimum altitude	2451 m	2424 m	1462 m	1503 m
Average altitude	1974 m	1975 m	1984 m	1973 m
Maximum altitude	1592 m	1478 m	1469 m	1438 m

Historical grey scale traditional aerial photos from 1954 (7 photograms) and 1991 (4 photograms) were first acquired by high resolution scanning (1200 dpi) and then geometrically orthorectified and co-registered on the basis of a local high resolution Digital Elevation Model (DEM), having a 40 m resolution, and a number of Ground Control Points (GCP) for each photogram ranging between 46 and 80. The resulting average Root Mean Square Error (RMSE) of the orthorectification and co-registration was equal to 0.3 m for the photograms from 1954 and to 0.7 m for the photograms from 1991. The spatial distribution of the mountain pine at the year 2007 was created by visual interpretation and manual delineation in a GIS (Geographic Information System) environment of the 2007 orthophoto. A nominal scale of 1:5000 was adopted; pine formations were mapped when reaching minimum crown coverage of 10% and a minimum size (minimum mapping unit) of 500 m². Each polygon was classified according to crown cover classes: I < 40%; II between 40 and 80%; III > 80%. Following the procedure detailed in Chirici et al. (2006), the 2007 map was then modified on the basis of the 1999, 1991 and 1954 orthophotos, in order to obtain the historical mountain pine distribution maps at the different years (Fig. 2.2). All the maps were then overlayed with the DEM in order to extract the average, minimum and maximum altitude of mountain pine distribution at the four different investigated dates.

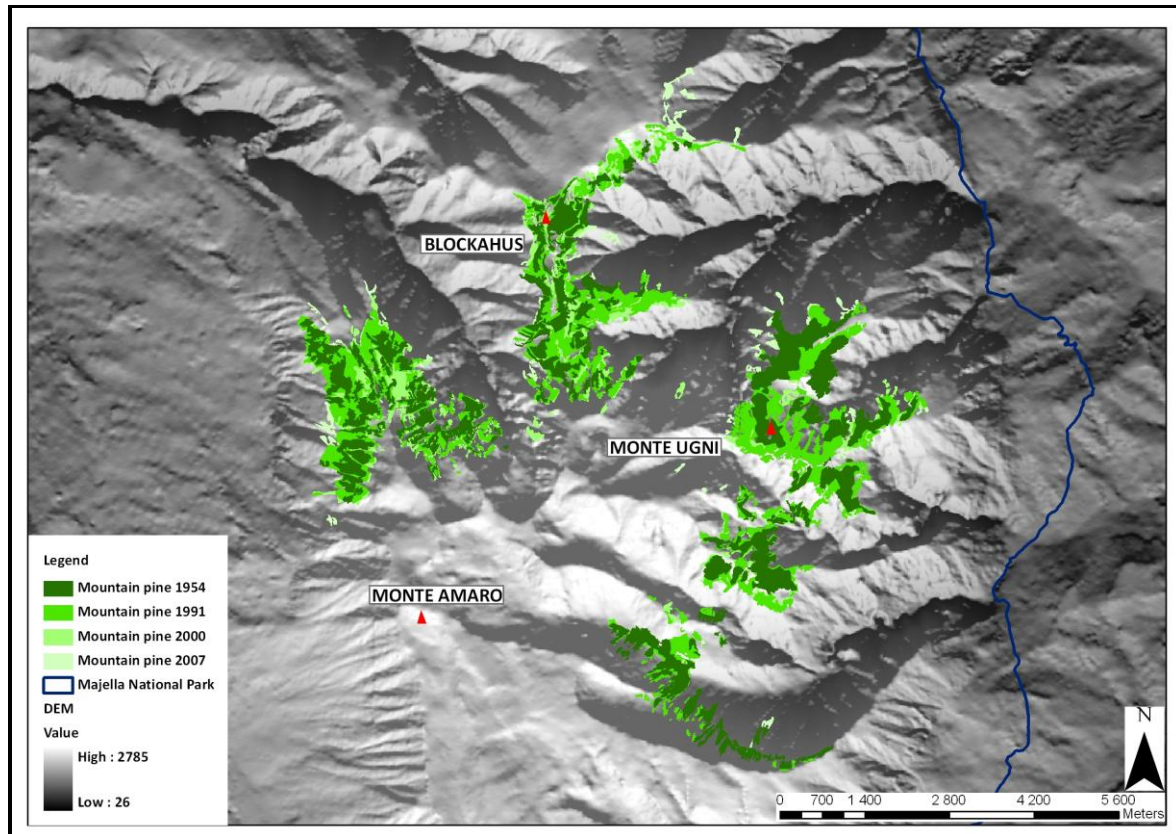


Fig. 2.2 Multitemporal maps of the mountain pine geographical distribution on the basis of the digital elevation model.

The spatial distribution of the mountain pine forest in the test area changed from 693 ha in 1954 (4,8% of the study area) to 1423 ha in 2007 (9,8% of the study area) (Table 2.1). At the last investigated date (2007) the altitudinal range of the mountain pine distribution ranges between 1438 and 2503 m a.s.l., with an average elevation of 1973 m. During the last 52 years, the mountain pine forest expanded 52 m upward and 154 m downwards. Maximum forest elevation changed since 1954 (2451 m), to 1991 (2424 m), to 1999 (2462), until 2007 (2503 m), with an overall increase of about 1 m per year (Table 2.1). Likewise, minimum forest elevation has been constantly decreasing since 1954 (1592 m), to 1991 (1478 m), to 1999 (1469), until 2007 (1438 m); about 3 m per year (Table 2.1). Overall, forest cover in the study area has increased yearly at higher rate in the period 1954-1991 (2.1%), followed by a period of partial decrease in rate, 1991-1999 (1.8%), and then a period of stable rate, 1999-2007 (0.4%) (Fig. 2.3).

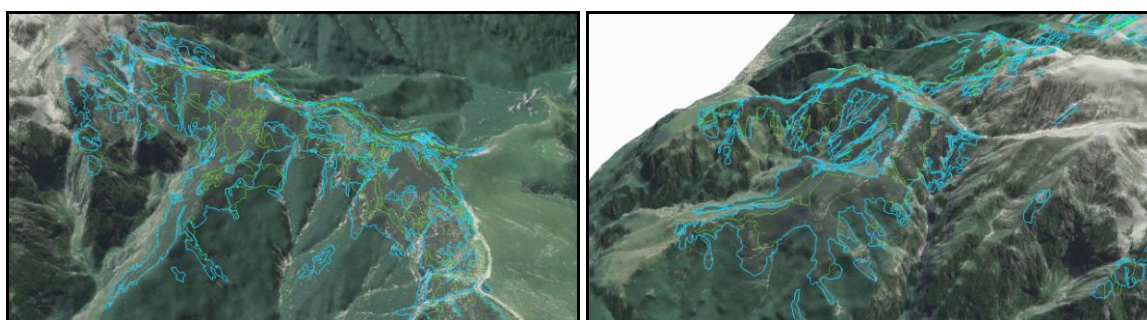


Fig. 2.3 3D ortophotos taken in 2007; mountain pine forest contour in 1954 (green) and 2007 (blue). View of M. Blockhaus (upper picture) and of M. Ugni (lower picture).

2.4 Canopy cover and grazing history in the Majella National Park

The grazing history was assessed for the last two hundred years for the provinces within the borders of the Majella National Park, and for municipalities (villages) within the study area, using archival documents (official statistics, historical archives of municipalities and provinces). The forest cover versus time course provides a proxy indicator for reconstructing forest cover change, whereas the number of sheep versus time allows an indirect assessment of grazing rate changes.

Sheep comprised more than 80% of the total livestock in the municipalities of the study area (such a percentage decreasing more recently), with land use widely dedicated to livestock rearing, according to historical records. Archival data, though discontinuous, are available from these municipalities (and local sources) to historically document livestock variation. Grazing intensity peaked in the middle of the 19th century, decreasing over the last century (Table 2.2). A first sharp reduction in grazing intensity took place in the first half of the 20th century and a second one in the last 30 years. During the first decade of present century, pasturing livestock has become less and less important.

Mountain pine forest cover has increased during the past 50 years with a mean annual increment of 13.7 ha. Forest cover was almost proportionally distributed across the three crown cover classes, when grazing was moderately intense, from early 1920s to late 1950s (Table 2.2).

Table 2.2 Tree establishment at the tree line and main land-use changes in the study area. Forest cover classes are: I, < 40%; II, between 40 and 80%; III, > 80%. Transhumance, long-distance movement; pastoralism, short-distance migration. *Averaged population for the municipalities of the Majella National Park.

Provinces of Pescara – Chieti – L’Aquila					
Year	Heavy grazing (transhumance and pastoralism) and cutting	Moderate grazing (pastoralism) and cutting	Low grazing (pastoralism) and cutting	Mountain pine forest cover (ha)	Forest cover class (% of total)
1880	Sheep n. > 500000 Inhabitant n. 800000 (126620*)				
1890					
1900					
1910					
1920		300000 < Sheep n. <500000 Inhabitant n. 943000 (125585*)			
1930					
1940					
1950				685 (1954)	I, 24%; II, 37%; III, 39%
1960					
1970					
1980			Sheep n. < 300000 Inhabitant n. 978000 (92521*)		
1990				1196	I, 6%; II, 7%; III, 87%
2000				1327	I, 13%; II, 22%; III, 65%
2010				1409 (2006)	I, 13%; II, 23%; III, 64%
Municipalities of the Majella National Park					
1982			30116		
1990			41143		
2000			31352		
2005			17769		
2008			23081		
2009			14715		

More recently, forest cover has become increasingly skewed towards class III. The highest number of inhabitants in the municipalities of the Majella National Park was in the beginning of the 20th century. Afterwards, the number of inhabitants began to decrease gradually up to the beginning of present

century, when the population statistics recorded a plateau. The population of the larger area (provinces of Pescara, Chieti, L'Aquila), instead, has increased gradually through the last 100 years, following a shift from agriculturally-based to tertiary sector economy.

2.5 Land use change and mountain pine forest dynamics

The increase of approximately 1 K in average annual air temperature in the last 100 years all over Italy (Brunetti et al. 2006), would point to tree recruitment above the present forest limit. Our results show that the forest–pasture ecotone of the Majella massif has changed fundamentally during the last 50 years, involving encroachment processes, which is consistent with the substantial changes in land use in those ecosystems. Neither increasing growth trends nor decreasing ages in mountain pine along an elevation transect were observed by Palombo (2013), pointing to a minor role played by climate in shaping the current tree line on the Majella massif.

In an analysis of recent forest dynamics along an altitudinal gradient in the upper Susa Valley (western Italian Alps), Motta et al. (2006) argued that tree establishment was mainly controlled by land use, while tree growth was for the most part controlled by climate. Nevertheless, recent establishment above current tree lines in response to climate warming has been found in many regions of the Northern Hemisphere (e.g. Daly and Shankman 1985; Kullman 1991; Luckman and Kavanagh 1998). Studies in the Swiss Alps showed a dramatic increase in the growth of *Picea* and *Pinus* at the tree line (Paulsen et al. 2000) and a dramatic increase of Scots pine at its southern distribution limits (Giuggiola et al. 2010). Rising temperatures have increased the productivity and survival of trees in mountain environments (Paulsen et al. 2000; Motta and Nola 2001), with a resulting expansion of woodland mosaic in the Alps (Chauchard et al. 2007). In Mediterranean mountains, instead, patterns of land abandonment and livestock grazing superimpose to the effect of climate change, and therefore these observations have to be reframed in a context of land-use change (Poyatos et al. 2003, Boden et al. 2010).

On the Majella massif, abundant mountain pine regeneration was observed during the past 50 years (Fig. 2.4), simultaneously with abandonment of summer pastoralism above the beech tree-line (van Gils et al. 2012) and climate becoming drier during the growing season (Palombo 2013). The interpretation of historic photographs indicates that areas that are now covered by closed mountain pine forest were covered by scattered vegetation in the mid of the 20th century. The considerable expansion of mountain pine in large areas of the Majella plateau, which have been grazed or cut until recently, is probably due to the abandonment of human activities in these marginal lands.

At these tree lines, cessation of grazing has been relatively gradual and marked by a staggered pattern, and forest expansion has not occurred homogeneously at the local scale. More interesting, however, are the even larger intrusion and colonization of mountain pine downhill in crop systems and beech forests. These results agree with several studies carried out recently in different European mountains (Švajda et al. 2011; Mihai et al. 2007; Jodłowski 2006; Mind'áš et al. 2004), in which the authors attribute the recent expansion of mountain pine *krummholz* to its ability to recolonize site from previous periods. In addition, Gerigh-Fasel et al. (2007) establish that the altitude of the potential regional tree line (derived from the highest forest patches identified by a moving window algorithm) as a reference to discern which is the main factor influencing the upward expansion of the tree line. Upward shifts above the potential regional tree line were considered to be influenced primarily by climate change, while upward shifts below the potential regional tree line were interpreted as primarily influenced by land abandonment. If we consider these claims and that mountain pine could reach the summit of the Majella massif (Migliaccio 1966), we could affirm that land use change was the main factor that led the rapid expansion of mountain pine forest, through the re-colonization of abandoned pastures. This expansion occurred in the altitudinal range of mountain pine distribution and towards both higher and lower altitudes. On the other hand, also climate variables may affect tree growth at the high mountain pine limit, by reacting areas close to the local tree line in subalpine pastures grasslands, which are very resistant against invasion of tree species (Gerigh-Fasel et al. 2007).

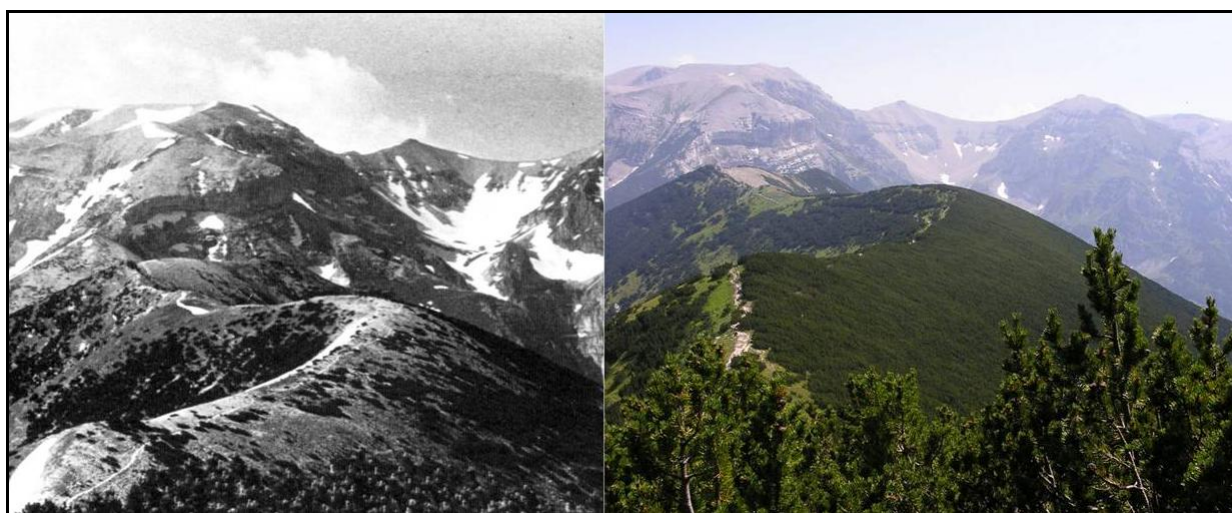


Fig. 2.4 Mountain pine dominated tree line at the Majella National Park (Mt. Cavallo), Central Apennines, Italy; left photo taken in 1940, right photo taken in 2005. The most evident change is the increase in tree density across the tree-line ecotone together with an upward shift of several meters in some areas.

Several studies report on dendroclimatic elements on mountain pine at tree line (Schueller and Rolland 1995; Rolland et al. 1998). Pelfini et al. (2006), for mountain pine in Central Italian Alps,

found that the climate of the summer months has the strongest influence on tree-ring growth. Correlations between ring widths and climatic conditions on mountain pine populations of the Majella massif (Di Cosmo 2003; Palombo 2013) suggest that warmer spring temperatures would be helpful to expand the vegetation period. Low temperature and abundant snowfall during spring delay bud burst, reducing growth time span, while warmer air induces prompt shoot development.

In Mediterranean mountains, constraints on tree-line dynamics probably include processes more contingent on local environmental conditions than on the recent disturbance history (Batllori et al. 2010); indeed recruitment has not been episodic in the studied mountain pine stands. A small change in site-specific water availability could have a profound influence on stand composition, structure, and function, in Mediterranean climate (Joffre and Rambal 2002; Vitale et al. 2012). Despite the relatively high annual precipitation in the study area, regular and prolonged summer droughts point to the Mediterranean mark of this forest ecosystem. The high solar radiation coupled with high soil permeability due to karst and fissuring of limestone, may result in high evaporative water loss and low water holding capacity (Anfodillo et al. 1998; Boden et al 2010). Dirnböck et al. (2003) suggested a considerable range expansion of mountain pine at only moderate levels of temperature increase, throughout most of the north-eastern calcareous Alps. Overall, our results indicate that, on the Majella massif, past seasonal livestock residence due to shepherds' tracks and transhumance might be stimulating growth more than rising temperature.

The recent beech forest regeneration and expansion at the lower limit of mountain pine forests would depend on canopy openings and temperature trends (Vitale et al. 2012) and would be delayed only in the presence of disturbance (i.e. forest fire). The rare beech forest fire in Majella can be attributed to a combination of the several conditions, including summer drought and abandoned farmland management (van Gils et al. 2010). Instead, on the Majella massif, van Gils et al. (2008) observed that from 1975 to 2003 beech advanced into abandoned farmland and subalpine pastures from the contiguous, mid-altitudinal beech forest and from beech outliers, at a rate of 1.2% per year. The same applies to the mountain pine ecotone in other European mountains, where the on-going reduction of krummholz surface is the result of the upward shift of spruce forest (Mihai et al. 2007; Mind'áš et al. 2004). Mediterranean mountains will probably present shifts and replacements in response to climate and land-use changes, as a result of interfering mechanisms between different species (Lingua et al. 2008).

2.6 Conclusions and perspectives

The forest–pasture ecotone of the Majella massif has changed markedly during the last 50 years. A major role was played by changes in land use, i.e. decreasing human pressure (animal husbandry and tree cutting). The expansion towards lower altitude of mountain pine and the possible interaction with beech forest remain to be investigated. Evidence of rapid alterations due to land-use modifications in the tree line of Majella massif provides additional indication of complex effects of global change on mountain ecosystems facing the Mediterranean Basin (cf. Améztegui et al. 2010; Chauchard et al. 2010), and shows how difficult is to disentangle complex processes, such as the impact of global change on mountain ecosystems. Climate change is altering biological processes and having important impacts on biodiversity at multiple scales. However, the responses of species and biological communities to climate change can also be influenced by the additive or synergistic effects of other components of global change, such as land use changes or biological invasions. In fact, the magnitude of the impacts of each one of the different components of global change, and therefore their interactions, is subjected to variation among systems and biomes (Sala et al. 2000; Parmesan 2006; Clavero et al. 2011).

In a recent study, Bracchetti et al. (2012) confirmed the crucial role of the abandonment of rural and mountain areas, which induced changes both at landscape level and at plant community level. The most common and widespread effect has been either the expansion of shrublands or forests, or both, as observed on the Mediterranean mountains. They also highlighted that the disappearance of Mediterranean cultural landscapes is a relevant problem for biodiversity conservation, causing the disappearance of a large number of species, habitats and landscapes in the Mediterranean Basin. The cultural landscape of the Mediterranean region is the result of the millenarian integration between land use and natural processes (cf. Finck et al. 2002; Mazzoleni et al. 2004).

Others studies conducted within the Mediterranean Basin, however, analysed the effect of climate changes on mountain biodiversity, and forecasted their effects on the distribution and community composition of tree species. The main conclusion were that areas with climatic conditions suitable for cold-adapted species will decrease significantly under climate change (cf. Gottfried et al. 2012) and that Mediterranean mountains might lose their key role as refugia for cold-adapted species and thus an important part of their genetic heritage (Alkemade et al. 2011; Ruiz-Labourdette et al. 2012). In this context Resco de Dios et al. (2007) suggested an active forest management to curb global warming effects, divided into three approaches: i) Conservation Management Strategy, which aims to prevent emissions and conserve current forest carbon pools through diminishing deforestation, increasing rotation period, reducing thinning intensity and restricting many harvesting activities; ii) Storage Management Strategy, which aims to increase the amount of carbon stored in vegetation

and soil through an increase in forested areas and/or carbon stored per unit area in natural and plantation forests; iii) Substitution Management Strategy, which includes the transfer of carbon biomass into furnishings, construction and other. Priorities of these management strategies are the promotion and conservation of biological diversity. This is achieved by: i) sustainable forest management for a preservation of the pool of natural species and genetic variability of forests, and ii) networking protected areas or natural ecosystems by creating ecological corridors or by maintaining appropriate ecological components in associated areas to allow natural migration of ecosystem elements. In protected areas such a National Park, is fundamental to describe the best management practices that will maintain or enhance adaptive capacity or resilience of ecosystems. In some cases, the Authorities may need to reconsider the current position, which encourage the decline of traditional practices (e.g. seasonal grazing), to avoid the risk of losing very rare ecosystems (Bartolomé et al. 2008). The elaboration of techniques for reading the landscape and its assets that can help in identifying and cataloguing landscape units, as well as in recognizing their ecologic values, and in creating models inspired to a sustainable territorial development, should be reconsidered. Indeed, National Park zonation considers, in general, various areas with different levels of protection, including core areas, buffer zones, and transition zones. Restriction in land use systems is relatively clear for core areas and transition zone, while many definitions of buffer zones can be provided. Zonation on national parks in mountain areas of the Mediterranean region, for instance, should be designed to help achieve the conservation needs of the park, basing the analysis of buffer zones (e.g. the contiguous area between beech and mountain pine forests on the Majella massif) on the accomplishment of conservation needs.

The question of how to meet wood demand at the least cost to forest biodiversity in rural landscapes of Mediterranean mountains could require the evaluation of two contrasting alternatives: sustainable forest management, which integrates both objectives on the same area; and strict forest conservation, in which high-impact forestry is combined with protecting high-value forest stands. While separating land for forest conservation from land for forest management could be fundamental to reducing emissions from deforestation and forest degradation (REDD), requiring the sustainable intensification of forestry, sustainable forest management is often an aim of certification schemes, and can result from some forms of agroforestry.

Acknowledgements We thank the Majella National Park for access to sampling sites and permission to core trees. Financial support for Caterina Palombo was provided by a Majella National Park fellowship. We are grateful to Angela Stanisci, Paolo Di Martino, Bruno Lasserre, Elena Liberatoscioli, Giovanni Pelino, Edoardo Micati, and Hein van Gils for valuable suggestion and fruitful discussion.

References

- Agnoletti M. 2007. The degradation of traditional landscape in a mountain area of Tuscany during the 19th and 20th centuries: Implications for biodiversity and sustainable management. *For Ecol Manage* 249: 5-17.
- Alkemade R., Bakkenes M., Eickhout B. 2011. Towards a general relationship between climate change and biodiversity: an example for plant species in Europe. *Reg Env. Ch.* 11: 143-150.
- Améztegui A., Brotons L., Coll L. 2010. Land-use changes as major drivers of mountain pine (*Pinus uncinata* Ram.) expansion in the Pyrenees. *Global Ecol Biogeogr* 19: 632-641.
- Anfodillo T., Rento S., Carraro V., Furlanetto L., Urbinati C., Carrer M. 1998. Tree water relations and climatic variations at the alpine timberline: seasonal changes of sap flux and xylem water potential in *Larix decidua* Miller, *Picea abies* (L.) Karst and *Pinus cembra* L. *Ann For Sci* 55: 159-172.
- Bartolomé J., Boada M., Saurí D. 2008. Conifer Dispersion on Subalpine Pastures in Northeastern Spain: Characteristics and Implications for Rangeland Management. *Range Ecol Manage* 61: 218-225.
- Batllo E., Camarero J.J., Gutiérrez E. 2010. Current regeneration patterns at the tree line in the Pyrenees indicate similar recruitment processes irrespective of the past disturbance regime. *J Biogeogr* 37: 1938-1950.
- Blasi C., Di Pietro R., Fortini P., Catonica C. 2003. The main plant community types of the alpine belt of the Apennine chain. *Plant Biosyst* 137: 83-110.
- Blasi C., Di Pietro R., Pelino G. 2005. The vegetation of alpine belt karst-tectonic basins in the Central Apennines. *Plant Biosyst* 139: 357-385.
- Blondel J. and Aronson J. 1999. Biology and wildlife of the Mediterranean region. Oxford University Press, Oxford.
- Boden S., Pyttel P., Eastaugh C.S. 2010. Impacts of climate change on the establishment, distribution, growth and mortality of Swiss stone pine (*Pinus cembra* L.). *iForest* 3: 82-85.
- Bracchetti L., Carotenuto L., Catorci A. 2012. Land-cover changes in a remote area of central Apennines (Italy) and management directions. *Land Urb Plan* 104: 157-170.
- Brunetti M., Maugeri M., Monti F., Nanni T. 2006. Temperature and precipitation variability in Italy in the last two centuries from homogenised instrumental time series. *Int J Clim* 26: 345-381.
- Chauchard S., Carcaillet C., Guibal F. 2007. Patterns of land-use abandonment control tree-recruitment and forest dynamics in Mediterranean mountains. *Ecosystems* 10: 936-948.
- Chauchard S., Beilhe F., Denis N., Carcaillet C. 2010. An increase in the upper tree-limit of silver fir (*Abies alba* Mill.) in the Alps since the mid-20th century: A land-use change phenomenon. *For Ecol Manage* 259: 1406-1415.

- Chirici G., Corona P., Koehl M. 2006. Earth observation techniques and GIS as tools for assessing land use/cover changes in a landscape context. In *The conservation of cultural landscapes*, Agnoletti M. (ed.). CAB International, Wallingford, pp. 57-70.
- Clavero M., Villero D., Brotons L. 2011. Climate change or land use dynamics: do we know what climate change indicators indicate? *PLoS ONE* 6: e18581.
- Dai Li 2010. Tree line change in Majella, Italy: trends, causes and predictions [Thesis]. The Netherlands: International Institute for Geo-information Science and Earth Observation.
- Daly C. and Shankman D. 1985. Seedling establishment by conifers above tree limit on Niwot Ridge, Front Range, Colorado, USA. *Arct Alp Res* 17: 389-400.
- Debussche M., Lepart J., Dervieux A. 1999. Mediterranean landscape changes: evidence from old postcards. *Global Ecol Biogeogr Lett* 8: 3-15.
- Di Cosmo L. 2003. Considerazioni sull'esistenza delle annate di pasciona nel pino mugo mediante l'analisi dendroecologica in una stazione della Majella. *Ita For Mont* 58: 173-190.
- Dirnböck T., Dullinger S., Grabherr G. 2003. A regional impact assessment of climate and land-use change on alpine vegetation. *J Biogeogr* 30: 401-417.
- Finck P., Riecken U., Schröder E. 2002. Pasture landscapes and nature conservation. New strategies for preservation of open landscapes in Europe. In *Pasture landscapes and nature conservation*, Redecker B., Finck P., Härdtle W., Riecken U. and Schröder E. (eds.). Berlin: Springer, pp. 1-14.
- Gehrig-Fasel J., Guisan A., Zimmermann N.E. 2007. Treeline shifts in the Swiss Alps: Climate change or land abandonment? *J Veg Sci* 18: 571-582.
- Giuggiola A., Kuster T.M., Saha S. 2010. Drought-induced mortality of Scots pines at the southern limits of its distribution in Europe: causes and consequences. *iForest* 3:95-97.
- Gottfried M., Pauli H., Futschik A., Akhalkatsi M., Barančok P., Benito Alonso J.L., Coldea G., Dick J., Erschbamer B., Fernández Calzado M.R., Kazakis G., Krajči J., Larsson P., Mallaun M., Michelsen O., Moiseev D., Moiseev P., Molau U., Merzouki A., Nagy L., Nakhutsrishvili G., Pedersen B., Pelino G., Puskas M., Rossi G., Stanisci A., Theurillat J.P., Tomaselli M., Villar L., Vittoz P., Vogiatzakis I., Grabherr G. 2012. Continent-wide response of mountain vegetation to climate change. *Nature Climate Change* 2:111-115.
- Holtmeier F.K. 1981. What does the term 'krummholz' really mean? Observations with special reference to the Alps and the Colorado Front Range. *Mount Res Dev* 1: 253-260.
- Jodłowski M. 2006. Geographical controls on the course of the upper mountain pine (*Pinus mugo*) limit in the Tatra mts. *Ekologia* 25: 105-114.
- Joffre R. and Rambal S. 2002. Mediterranean ecosystems. Encyclopedia of Life Sciences. Macmillan Publishers Ltd, London.

- Kohler T., Pratt J., Debarbieux B., Balsiger J., Rudaz G., Maselli D. 2012. Sustainable Mountain Development, Green Economy and Institutions. From Rio 1992 to Rio 2012 and beyond. *Final Draft for Rio 2012*.
- Kullman L. 1991. Structural change in a subalpine birch woodland in north Sweden during the past century. *J Biogeogr* 18: 53-62.
- Lingua E., Cherubini P., Motta R., Nola P. 2008. Spatial structure along an altitudinal gradient in the Italian central Alps suggests competition and facilitation among coniferous species. *J Veg Sci* 19: 425-436.
- Luckman B.H. and Kavanagh T.A.. 1998. Documenting the effects of recent climate change at treeline in the Canadian Rockies. In *The impacts of climate change on forests. Lecture Notes in Earth Sciences*, : Beniston M. and Innes J.L. (eds.). Berlin: Springer Verlag, pp. 121-144.
- Mazzoleni S., Di Pasquale G., Mulligan M., Di Martino P., Rego F. 2004. Recent dynamics of the mediterranean vegetation and landscape. Chichester, UK: Wiley & Sons Ltd.
- Migliaccio F. 1966. La vegetazione a *Pinus pumilio* della Majella. *Ann Bot (Roma)* 28:539–550.
- Mihai B., Savulescu I., Sandric I. 2007. Change detection analysis (1986–2002) of vegetation cover in Romania. *Mount Res Dev* 27: 250-258.
- Mind'áš J., Čaboun V., Priwitzer T. 2004. Timber line and expected climate changes [in Slovak]. Kadlečík J. (ed.). Year-book Turiec and Fatra. Vrútky, Slovakia: ŠOP SR, pp. 17–23.
- Motta R. and Nola P. 2001. Growth trends and dynamics in subalpine forest stands in the Varaita Valley (Piedmont, Italy) and their relationships with human activities and global change. *J Veg Sci* 12: 219-230.
- Motta R., Morales M., Nola P. 2006. Human land-use, forest dynamics and tree growth at the treeline in the Western Italian Alps. *Ann For Sci* 63: 739-747.
- Parmesan C. 2006. Ecological and evolutionary responses to recent climate change. *Annu Rev Ecol Evol Syst* 37: 637–669.
- Palombo C. 2013. The influence of land-use and climatic changes on mountain pine (*Pinus mugo* Turra spp. *mugo*) ecotone dynamics at its southern range margin on the Majella massif, Central Apennines [Dissertation]. Campobasso: University of Molise.
- Paulsen J., Weber U.M., Körner C. 2000. Tree growth near treeline: abrupt or gradual reduction with altitude? *Arct Antarct Alp Res* 32: 14-20.
- Pelfini M., Leonelli G., Santilli M. 2006. Climatic and environmental influences on mountain pine (*Pinus montana* Miller) growth in the central Italian Alps. *Arct Antarct Alp Res* 38: 614-623.
- Pisanelli A., Chiocchini F., Cherubini L., Lauteri M. 2012. Combining demographic and land-use dynamics with local communities perceptions for analyzing socio-ecological systems: a case study in a mountain area of Italy. *iForest* 5: 163-170.

- Poyatos R., Latron J., Llorens P. 2003. Land use and land cover change after agricultural abandonment, the case of a Mediterranean mountain area (Catalan pre-Pyrenees). *Mount Res Dev* 23: 362–368.
- Resco de Dios V., Fischer C., Colinas C. 2007. Climate Change Effects on Mediterranean Forests and Preventive Measures. *New For* 33: 29–40.
- Rolland C., Petitcolas V., Michalet R. 1998. Changes in radial tree growth for *Picea abies*, *Larix decidua*, *Pinus cembra* and *Pinus uncinata* near the alpine timberline since 1750. *Trees* 13: 40–53.
- Ruiz-Labourdette D., Nogués-Bravo D., Ollero H.S., Schmitz M.F., Pineda F.D. 2012. Forest composition in Mediterranean mountains is projected to shift along the entire elevational gradient under climate change. *J Biogeogr* 39: 162–176.
- Sala O.E., Chapin III F.S., Armesto J.J., Berlow E., Bloomfield J., et al. 2000. Global biodiversity scenarios for the year 2100. *Science* 287: 1770–1774.
- Schueller J.F. and Rolland C. 1995. Influence de l'altitude, de l'exposition et du climat sur la croissance du pin à crochets (*Pinus uncinata* Ram.) en Cerdagne (Pyrénées Orientales Françaises). *Pirineos* 145–146: 23–34.
- Stanisci A. 1997. Gli arbusteti altomontani dell'Appennino centrale e meridionale. *Fitosoc.* 34: 3–46.
- Stanisci A., Pelino G., Blasi C. 2005. Vascular plant diversity and global change in central Apennine (Italy). *Biodiv Cons* 14: 1301–1318.
- Švajda J., Solár J., Janiga M., Buliak M. 2011. Dwarf Pine (*Pinus mugo*) and Selected Abiotic Habitat Conditions in the Western Tatra Mountains. *Mount Res Dev* 31: 220–228.
- van Gils H., Batsukh O., Rossiter D., Munthali W., Liberatoscioli E. 2008. Forecasting the pattern and pace of *Fagus* forest expansion in Majella National Park, Italy. *App Veg Science* 11: 539–546.
- van Gils H., Odoi J.O., Andrisano T. 2010. From monospecific to mixed forest after fire? An early forecast for the montane belt of Majella, Italy. *For Ecol Manage* 259: 433–439.
- van Gils H., Conti F., Ciaschetti G., Westinga E. 2012. Fine resolution distribution modelling of endemics in Majella National Park, Central Italy. *Plant Biosyst* 146: 276–287.
- Vitale M., Mancini M., Matteucci G., Francesconi F., Valenti R., Attorre F. 2012. Model-based assessment of ecological adaptations of three forest tree species growing in Italy and impact on carbon and water balance at national scale under current and future climate scenarios. *iForest* 5: 235–246.

THIRD CHAPTER:

Warming-related growth responses at the southern limit distribution of mountain pine (*Pinus mugo* Turra ssp. *mugo*)

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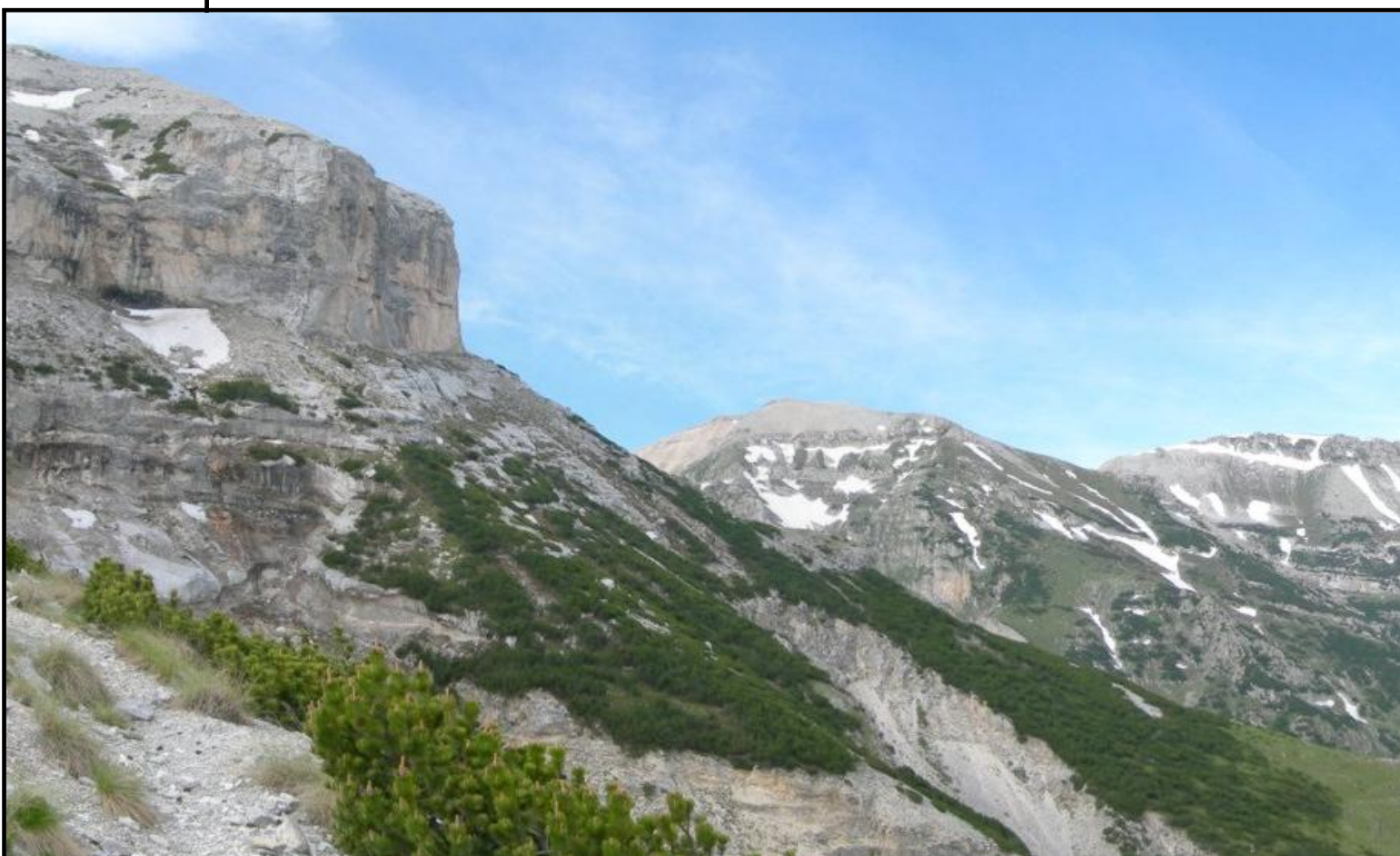
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Article accepted with revision by *Journal of Vegetation Science*



ABSTRACT

Questions: Could mountain pine (*Pinus mugo* Turra spp. *mugo*) located above the timberline and at the edge of its potential geographical distribution be a promising species to monitor climatic change in peculiar environments, such as the Mediterranean mountains? Does mountain pine krummholz upward expansion result from the current climate change? Have the relationship between climate and tree-ring growth of mountain pine changed? And, and to what extent does a climatic variability affects tree-ring growth? Could mountain pine upward expansion contribute to the loss of biodiversity in the undisturbed treeline ecotone on the Majella massif?

Location: Majella National Park, Apennines (Italy).

Methods: Dendroclimatological analysis was performed to examine the relationship between tree rings of mountain pine and climatic parameters at the tree line. Climate-growth relationships were analysed by correlation function (CF) analysis and moving CF (MCF).

Results: In the Majella massif, climate variables affected mountain pine tree growth at the treeline ecotone. Significant correlations were found with the spring maximum temperatures and summer precipitation. Essentially, three months played a key role in mountain pine growth: April, May and October of the previous year. MCF showed that mountain pine is a sensitive species, in terms of tree ring growth.

Conclusions: Growing above the limit of tree vegetation, mountain pine proved to be a promising species for monitoring climate- and land use-driven influences over tree ring growth at high elevation. Our results showed that mountain pine is very sensitive to climate, particularly at the beginning and at the end of the growing season.

3.1 Introduction

In the central Apennines (Italy), the lower altitudinal range and the strong influence of Mediterranean climate place the boundary between alpine and subalpine bioclimatic belts at 2550 m a.s.l. Within the Mediterranean basin, the Apennine range represents a temperate-montane climatic “island” within which the central Apennines forms an archipelago of arctic-alpine “islands” (van Gils et al. 2012). The Apennines chain acted as refugia for trees during the Würmian Lateglacial (Watson 1996), playing an important role in the preservation of an endemic high-altitude vegetation (Blasi et al. 2003). Future impacts of current climate change, such as the increase in air and surface temperatures and the variation in precipitations, may cause significant alterations in forest ecosystems, proportionally more perceptible at higher elevations (Boden et al. 2010). Therefore, on the Apennines it is very important to understand how these species could adapt their growth and distribution to the ongoing climate changes.

Mountain pine (*Pinus mugo* Turra ssp. *mugo*) is one of the rare and relict species of the Apennines. Although considered the potential natural vegetation at the subalpine belt in central Apennines (Migliaccio 1966), mountain pine forms pure populations only in the Abruzzo Region. The largest stands are located at the tree line ecotone on the Majella massif, particularly in the north-facing aspects, the southernmost limit of the distributional range of this species. Being the Majella massif the second highest pick of the Apennines (2793 m a.s.l., M. Amaro), the tree-line evolution of this mountain is probably climate-related. Mountain pine protects the soil with a strong potential against the erosion and in favour of water retention, especially on the slopes subject to landslides. It stabilizes the snow cover thus hampering the release of avalanches and provides habitat for many species of flora and fauna (Jodłowski 2006). Mountain pine krummholz (Holtmeier 1981) grows above the timberline, which in the North-central Apennines is formed for the most part by European beech (*Fagus sylvatica* L.) between 1.800–1.900 m a.s.l. (Piermattei et al. 2012).

Nicolussi and Patzelt (2006) described the alpine timberline as very sensitive to climatic variability. Hamilton (1999) and Beniston (2000) considered the altitudinal zonation of mountain vegetation and the biodiversity of these zones to be sensors that indicate climatic and environmental changes. Yet, tree populations located at the edge of their potential geographical distribution may respond strongly to climate change in comparison with those at the core of the range (Peñuelas et al. 2008; Linares et al. 2009; Carrer et al. 2010). All these variables influence the mountain pine growth, making it a promising indicator to understand the impact of climatic changes on Mediterranean mountains. On the Majella massif, the benefit of a spread oceanic regime could advantage beech woods and mountain pine krummholz (Stanisci et al. 2000; van Gils et al. 2008), causing a direct competition between these two species along the upper timberline. Hence the need to understand the interactions between these tree species in order to forecast a possible loss of mountain pine forest cover (Mihai et al. 2007), and to suggest future forest management strategies, separating areas designated to conservation from those committed to wood production or integrating the two issues on the same land.

Gottfried et al. (2012) studied all major European mountain systems (including the Majella massif), showing that current climate change gradually transforms mountain plant communities, mirroring the degree of recent warming. They also suggested a progressive decline of cold mountain habitats. An upward shift of forest tree species is predicted for the 21st century, as a function of climate scenarios, together with a massive reduction of plant diversity and a high community turnover (Thomas et al. 2004; Thuiller et al. 2005; Lenoir et al. 2008; Randin et al. 2009) or a modification of correlation between tree growth and climate (Carrer et al. 2007, 2010). Furthermore, a lengthening

of the growing season, particularly in temperature-limited environments such as high altitude and/or mid to high latitudes (Saxe et al. 2001; Grace et al. 2002; Menzel et al. 2006; Way 2011), seems to represent the response to recent climate change. In Mediterranean mountain ecosystems, however, a lack of studies at small spatial scales, which test a possible warming-induced upward shift of treeline ecotones, is observed (e.g., Camarero et al. 2006).

Change in land use intensity is another factor that may affect treeline shift (Améztegui et al. 2010). National parks are potentially ideal sites for research and monitoring challenging alternative management practices and conservation strategies. Forests in Europe have been spontaneously expanding into abandoned farmlands at annual rates of 0.7 to 1.4% (van Gils et al. 2008). In Italy, the rate over recent decades (1.1% per year) falls within this range, being associated with the post-World War II rural exodus (MacDonald et al. 2000; Nicolini et al. 2002). Planning and decision-making towards sustainable management of multifunctional Mediterranean mountain forest require further production and analyses of long-term scenarios of various forest aspects, including tree development and area fragmentation, based on environmental change assumptions and growth pattern reconstruction.

Several studies have focused on the analysis of the potential responses of mountain pine *krummholz* distribution to different future climatic and land use scenarios in mountain environments of major European ranges (Alps, Tatra, Carpathians, Pyrenees), because of its important role as subalpine species growing above the timberline. Most of these studies, through distribution analysis, demonstrated an upward expansion of mountain pine at European level (Gehrig-Fasel et al. 2007; Améztegui et al. 2010; Švajda et al. 2011; Mihai et al. 2012). This was also the case of Apennines and the Majella massif in particular (Palombo et al. in press). Regardless of the ecological role of mountain pine *krummholz*, it is important to demonstrate if this upward expansion results from the current climate change. In this context, it is crucial to understand if deviations in the response to recent climate change occur for mountain pine growth and to what extent these variations affects tree ring growth, in order to predict potentially negative interactions with rare and endemic alpine species, which may bring to loss of biodiversity in the undisturbed treeline ecotone on the Majella massif.

Dendroclimatic elements on mountain pine and related elevational gradients at the treeline ecotone are limited to alpine environments (Schueller and Rolland 1995; Rolland et al. 1998; Pelfini et al. 2006; Lingua et al. 2008). Our aim was to test a dendroclimatic approach to identify major climatic factors controlling the radial growth of mountain pine at the subalpine-alpine ecotone in the Majella massif and to assess the long term variation in tree sensitivity to climate. We hypothesized that

upward migration of mountain pines should have occurred in undisturbed treeline ecotones of central Apennines due to the recent warming.

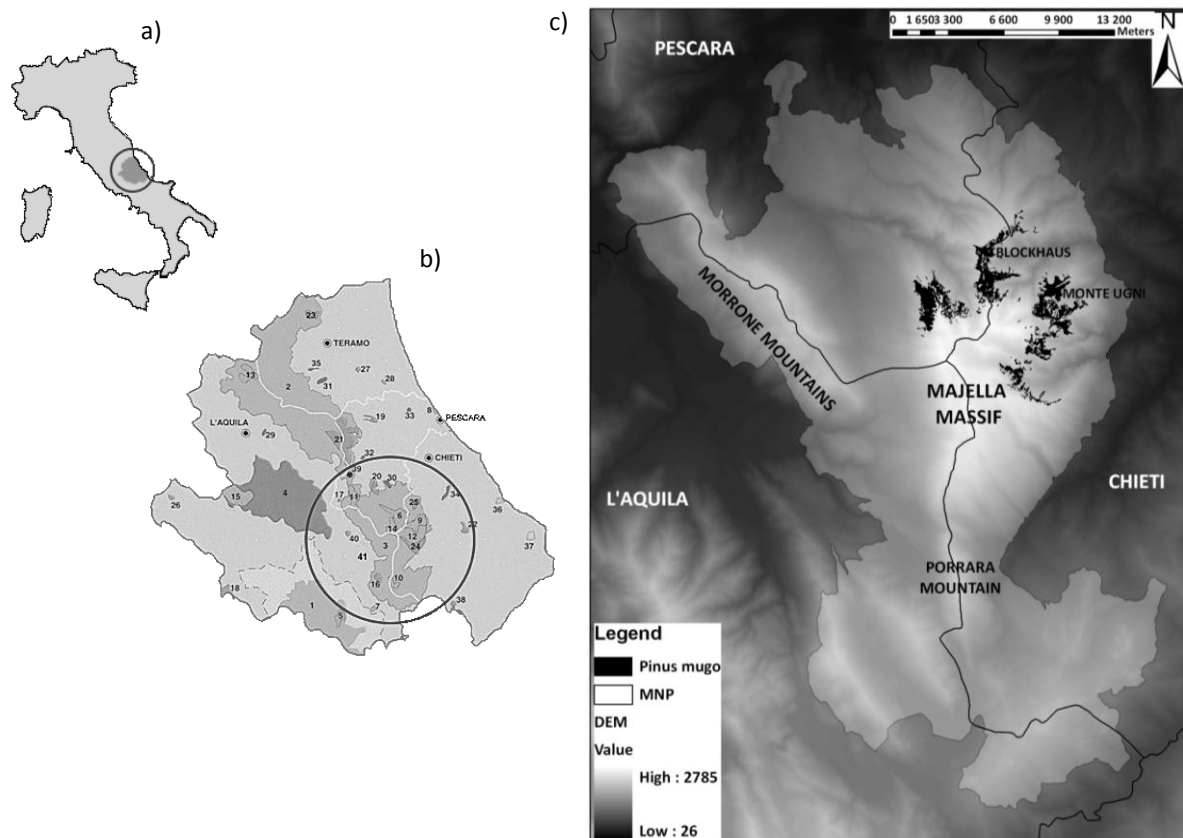


Fig. 3.1 (a) Map of the study region in the central Apennines, (b) representing the MNP and its location in Abruzzo (Italy); numbers indicate protected areas in Abruzzo. (c) Location of the study sites (Blockhaus, Monte Ugni) in the MNP, on the basis of the digital elevation model; in black the mountain pine distribution at 2007.

3.2 Materials and methods

3.2.1 Study site and sampling

The Majella massif represents the heart of the Majella National Park (MNP). Located in the Central-Apennines (Italy) (Fig. 3.1), MNP extends for 741 km² with an elevation ranging between approximately 130 and 2800 m a.s.l.. The landscape is dominated by NW–SE-oriented limestone ridges reaching above the beech tree-line at around 1750 m, i.e. Morrone, Rotella, Pizzalto and Porrarra and the calcareous Majella massif (van Gils et al. 2008, 2012). This latter reaches 2793 m a.s.l. with Mt. Amaro, creating a ridge with a latitudinal span between 42°09'33'' N and 42°00'14'' N, 32 km away from the Adriatic Sea. Here the typical rounded shape of peaks has led to the development of large plateau even at altitudes exceeding 2600 m a.s.l., forming a distinct

environment that is unique in the Apennine landscape. This high-altitude plateau collects the main vegetation features of the high elevation vegetation of Central Apennines, also enhanced by some peculiar floristic and coenological characteristics, which suggest a different background in the quaternary development of its vegetation (Blasi et al. 2005). The Majella massif is capped by a broad periglacial plain at around 2000 m a.s.l., including more than 15 smooth summits (Whitehead 1951; Stanisci et al. 2010), which was formed by the recurrent glaciations of the Apennines during the Pleistocene (Giraudi 2003) and the simultaneous retreats of the Adriatic sea (van Gils et al. 2012). The study area is located in the northern part of the Majella massif (Fig. 3.1), and corresponds to the subalpine-alpine humid type with mountain pine woodlands as potential natural vegetation slopes [habitat 4070 * bushes of *Pinus mugo* and *Rhododendron hirsutum* (Mugo-Rhododendretum hirsuti)] referring to the alliance *Epipactido atropurpureae*-*Pinion mugo* (Stanisci 1997). Mean annual temperature is 10.2 °C and total annual precipitation is 1140.2 mm, mainly concentrated in autumn and winter (Annex I, Fig. A1a). Mountain pine is a prostrate tree with a canopy height at adult stage varying between 0.3 and 2.5 m. It is a drought-tolerant species that grows successfully on sunny slope with carbonate-rich soils, but is very adaptable to poor and dry soils. In the study area, dense krummholz occupy the altitudinal (subalpine) zone between 1500 and 2300 m a.s.l., whereas small groups reach 2500 m a.s.l. (Palombo et al. in press), beyond the limit of forest growth.

In the late summer 2008, two sites were sampled on the opposite sides of the same valley. One site was located at Blockhaus (42°8'23.30" N – 14°06'40.24" E) east-facing, the other was located at M. Ugni (42°07'08.63" N – 14°09'19.81" E) west-facing. The two sites are located within a 4 km radius of each other and have the same altitudinal range (from 2000 to 2200 m a.s.l.). At each site, three study areas (30 x 20 m, Area 1 being the highest in elevation, Area 2 the intermediate and Area 3 the lowest) were defined at a distance of 10 m from each other, along a transect of 3000 m² (top-down, 30 x 100 m). In each study area 15-20 adult individuals of mountain pine were sampled for dendroecological and dendroclimatological analyses. Two wood cores were taken at 120° from each other with a 0.5-cm diameter increment borer. Trees were cored close to the base of the trunk and on the cross-slope side of the trunk to avoid sampling on the branches and compression wood. Plants had prostrate trunk, forming krummholz, which made irrelevant the collection of diameter at breast height and canopy height.

3.2.2 Tree-ring analysis

The tree cores were air dried, mounted on woody supports and sanded, following the standard dendrochronological procedures (Schweingruber 1988). Ring widths were measured with a 0.01 mm

resolution through LINTAB-measurement equipment coupled to a stereomicroscope (60× magnifications, Leica, Germany) and Time Series Analysis Programme (TSAP) software (Frank Rinn, Heidelberg, Germany). Raw ring-width chronologies of each dated tree were cross-dated first visually and then statistically using the software COFECHA (Holmes 1983). As a result of this process, 20% of the total cores were discarded, because of the presence of very irregular growth patterns, compression wood or missing rings.

The software ARSTAN (Cook and Holmes 1984), developed for processing tree-ring series from closed canopy, was used to standardize individual chronologies and produce raw and standardized mean chronologies for each study area. In order to emphasize higher interannual frequency variations, a spline function with a 50% frequency response of 20 years was fitted to each tree-ring raw series, computed by dividing observed by expected values. Cubic smoothing spline curves are the most suitable for removing non-climatic noises as long-term trend and effect of localized disturbance events that characterize natural forest dynamics, especially in a dense and close mountain pine shrubland. These events can lead an abrupt growth response (totally different from the typical decreasing trend of tree circumference, not available for mountain pine individuals) that can conceal the climatic signal even for many years after the disturbance event. The only risk of smoothing spline is to remove possible low-frequency climatic information (Cook and Peters 1981). For each study area, b-weight robust mean was used to develop a mean standardized chronology (STD). High correlation coefficients between standard mean chronologies of the two areas with the same altitude (see Results) allowed grouping all the chronologies in 3 altitudinal plots (Plot H the highest in elevation, Plot M the intermediate and Plot L the lowest)

Subsequently, a second cross dating between individual standard chronologies with the same altitude was carried out using COFECHA, discarding an additional 5% of the total cores to obtain mean chronologies with higher statistical accuracy. Three new standardized mean chronologies were obtained following the same steps with the ARSTAN program.

Such mean standard chronologies were used to analyse climate-growth relationship. Several descriptive statistics were used to compare key properties of each chronology for each study area (Table 3.1). These statistics included (1) the mean sensitivity (MS) and the tree-ring width standard deviation (SD), useful to assess the high-frequency variations; (2) the first order autocorrelation (AC) to detect the persistence retained before and after the standardization; (3) the mean correlation coefficient (\bar{r}) for all possible pairings of tree-ring series from individual cores over a common time interval (Briffa 1995) and (4) the expressed population signal (EPS) (Wigley et al. 1984) which was determined by calculating the chronology signal as a fraction of the total chronology variance.

EPS quantifies the degree to which a particular sample chronology portrays a hypothetically perfect chronology.

Table 3.1 Descriptive statistics for mountain pine tree-ring chronologies at Blockhaus and Monte Ugni. Area 1 is the highest in elevation, Area 3 being the lowest

	Blockhaus			M. Ugni		
Descriptive statistics	<i>Area 1</i>	<i>Area 2</i>	<i>Area 3</i>	<i>Area 1</i>	<i>Area 2</i>	<i>Area 3</i>
<i>Chronology time span (total years)</i>	1934-2007 (74)	1901-2007 (107)	1916-2007 (92)	1896-2008 (113)	1873-2008 (136)	1907-2008 (102)
<i>Raw mean ring-width (MRW)</i>	1.887	1.225	1.882	0.774	1.049	1.224
<i>Standard deviation (SD)</i>	1.235	0.638	1.116	0.479	0.617	0.612
<i>Mean sensitivity (MS)</i>	0.273	0.313	0.230	0.234	0.182	0.178
<i>First-order serial autocorrelation (AC)</i>	0.209	0.144	0.032	-0.010	-0.010	-0.005
<i>Mean inter-series correlation (rbar)</i>	0.271	0.222	0.259	0.179	0.170	0.097
<i>Expressed Population Signal (EPS)</i>	0.873	0.846	0.871	0.839	0.824	0.510

Note: MRW and SD were computed on the raw tree-ring series; MS, AC, rbar and EPS were computed on the indexed tree-ring series.

The same descriptive statistics were computed for the three mean standard chronologies of Plot H, Plot M and Plot L, in order to better compare their key properties (Table 3.2).

Table 3.2 Descriptive statistics for mountain pine tree-ring chronologies at different elevation. Plot H is the highest in elevation, Plot L the lowest.

Descriptive statistics	<i>Plot H</i>	<i>Plot M</i>	<i>Plot L</i>
<i>Chronology time span (total years)</i>	1896-2008 (113)	1880-2008 (129)	1886-2008 (123)
<i>Raw mean ring-width (MRW)</i>	1.041	1.206	1.654
<i>Standard deviation (SD)</i>	0.573	0.813	0.866
<i>Mean sensitivity (MS)</i>	0.230	0.201	0.181
<i>First-order serial autocorrelation (AC)</i>	- 0.021	- 0.014	- 0.001
<i>Mean inter-series correlation (rbar)</i>	0.187	0.225	0.205
<i>Expressed Population Signal (EPS)</i>	0.896	0.887	0.877

Note: MRW and SD were computed on the raw tree-ring series; MS, AC, rbar and EPS were computed on the indexed tree-ring series.

3.2.3 *Climate data*

Climate data collected from several meteorological stations, included in the MNP boundaries, were provided by the Servizio Idrografico e Mareografico of Pescara (Italy). An exhaustive analysis of this dataset revealed that almost all the stations are located below an altitude of 1000 m and rainfall series are always longer and more complete than temperature series, since all the stations were originally installed as rain gauge stations. Three of these stations were considered as the most representative for the study area, because of their more exhaustive series and their geographical position (highest in elevation and/or proximity to the study area). Data recorded from the three stations were used for a description of the local climatic conditions (Annex I).

Considering that for dendroclimatological analysis there is a lack of time series long enough to cover the time range of our dendrochronological series, we decided to use climatic data derived from the CRU TS 3.1 gridded dataset, with 0.5° of spatial resolution (Mitchell and Jones 2005). This choice was supported by significant correlations obtained between monthly observation from CRU dataset and monthly climatic data recorded by the three most representative stations of the study area (Annex I, Table A1b). We selected the averaged climatic data from the 4 grid points closest to the research area. For the period 1901-2009, maximum and minimum monthly temperature and total monthly precipitation were downloaded from the CRU TS 3.1 gridded dataset.

3.2.4 *Climate influence on tree growth*

Climate influence on tree growth was assessed using the bootRes package (Zang and Biondi 2012) in R environment. We examined the climate-growth relationship by correlation function (CF) analysis (Fritts 1976) and moving CF (MCF) for detecting the stationarity and consistency of CFs over time. bootRes applies a bootstrap process (Guiot 1991) to assess the statistical significance of the correlation coefficients. Climate data from March of the previous year (t-1) to October of the current year (t) served as independent variables (20), and the standard mean chronologies for the three altitudinal plots were used as dependent variables.

3.3 **Results**

3.3.1 *Tree-ring chronologies*

Descriptive statistics of the mean standard chronologies of each study area are listed in Table 3.1. Considering the highly significant ($P < 0.001$) correlation coefficients found between standard

chronologies of the study areas with the same altitude at the two sites ($r = 0.35$ with $n = 74$ for Areas 1; $r = 0.30$ with $n = 107$ for Areas 2 and $r = 0.61$ with $n = 92$ for Areas 3), a single mean chronology for each altitude were computed. Tree-ring series obtained from mature mountain pine trees spanned from 24 years to 120 years with a mean tree age (at coring height) of 68 years. Mean tree ring width was 1.301, ranging from a minimum value of 0.356 to a maximum value of 3.383. Mean sensitivity (MS) and tree-ring width standard deviation (SD) were 0.204 and 0.751, respectively. Descriptive statistics of the three mean chronologies (altitudinal derivation) (Table 3.2) were consistent with those of the single Area chronologies, with an increase of the EPS values. Dating tree rings was successful, as shown by the synchronous occurrence of pointer years (Fig. 3.2) - years in which particularly narrow or wide rings are formed - in the mean standard chronologies.

3.3.2 *Climate response patterns of tree growth*

Values of correlation analysis between standard mean chronologies of the three altitudinal plots, monthly data of maximum and minimum temperature, and total precipitation are represented in Fig. 3.3. The correlation coefficients showed that both maximum temperature and precipitation were sensitive factors for tree-ring growth. The positive effect of the spring maximum temperature in radial growth delayed in time according to altitude, starting from March for Plot L (the lowest), moving to April for Plot M and to May at high elevation for Plot H. Maximum temperature affected radial growth, especially during the start of the growing season: significant correlations were found with March, April and May for Plot L, with April and May for Plot M, and with May for Plot H. Maximum temperatures in October of the previous year positively influenced radial growth of trees at all altitude. In addition, a positive effect of the maximum temperature of the previous year on tree growth was observed for Plot H (July), Plot M (August) and Plot L (April).

Minimum temperatures also had an important effect on tree growth, especially for Plot L, where significant correlations were found with April (t and $t-1$) and May, while for Plot H only the minimum temperature of May influenced the tree growth. Minimum temperatures of January and February correlated negatively with tree growth for Plot M, while minimum temperatures of December correlated positively for Plot L.

In the study area, the amount of precipitation showed an effect on the mountain pine growth: summer precipitation of June and July positively influenced radial growth of trees at all altitude (except July for Plot M), while precipitation of April affected negatively tree growth only for Plot L. Precipitation of the previous year correlated negatively with tree growth of Plot H (October), Plot M (June and August), and Plot L (August).

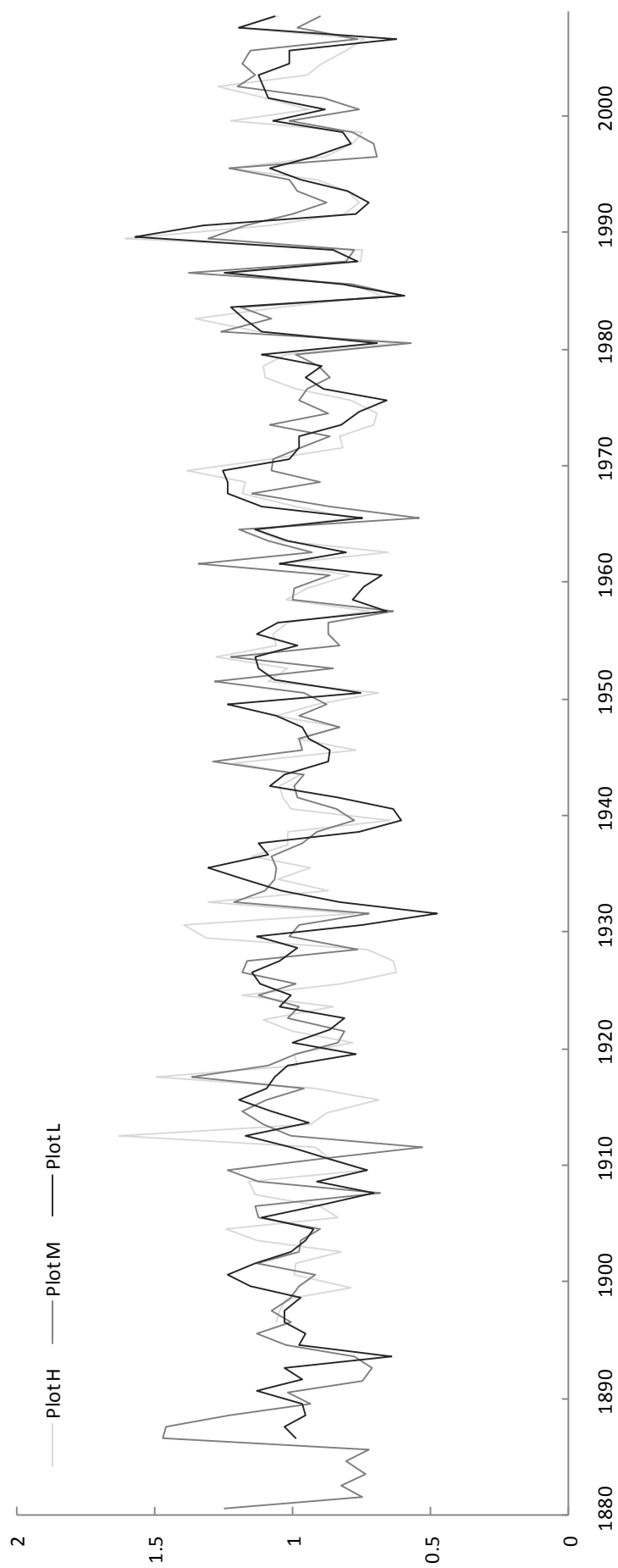


Fig. 3.2 Mean indexed chronologies for the three plots (Plot H, Plot M, Plot L) of mountain pine trees on the Majella massif.

Moving correlation functions (MCF, Fig. 3.4) confirmed the previous results and added a dynamic layout of tree responses to climate. All the significant CF variables exhibited the same response over time, only shifting significance, confirming the high sensitivity of tree growth to precipitation. Others CF non-significant variables were highlighted by MCF, e.g. with annual total precipitation of May and October (t and $t-1$) and current September, which showed significant transient response over time. Besides shifting significance, they also changed from negative correlation to positive correlation or vice versa, following the increasing or decreasing trend of total precipitation (Fig. 3.5). The same occurred for maximum and minimum temperature, where non-significant CF variables showed a step-wise shift in the variability of the climate response that highlighted tree ring growth sensitivity to temperature, exhibiting opposite but regular trend over time [e.g. February and April in Plot H, and October ($t-1$) in Plot L].

In particular:

- Precipitation of October showed a direct effect on the CF by the deep decrease during the period 1950-1990 (Fig. 3.5), causing a reversal of correlation coefficients from positive to significant negative during the current year.
- Climatic variables of April affected pine growth in different ways. In this case, a strong interaction between temperature and precipitation, and an evident difference of the correlation coefficients along the altitude, occurred (Figs. 3.4 and 3.5). Plot L was negatively affected by precipitation, while it was positively affected by maximum and minimum temperature during the whole time, despite of the clear and substantial fluctuations of climatic variables. The same happened in Plot M, where the correlation coefficients were less significant. In Plot H, on the contrary, the variable trend of climatic parameters affected tree-ring growth with an inversion of the correlations, particularly in maximum temperature, where correlation coefficients changed from significant negative to significant positive. The transition occurred gradually during the period 1930-1970, when temperatures were still decreasing but precipitation started to increase. Significant positive correlation coefficients appeared when precipitation showed a sharp increase and maximum temperatures started to increase (both around 1950-1990).
- During the same period, precipitation of May deeply decreased, causing a significant negative correlation in Plot L and Plot M, while temperatures that were already increasing reflected a significant positive correlation with tree-ring growth.

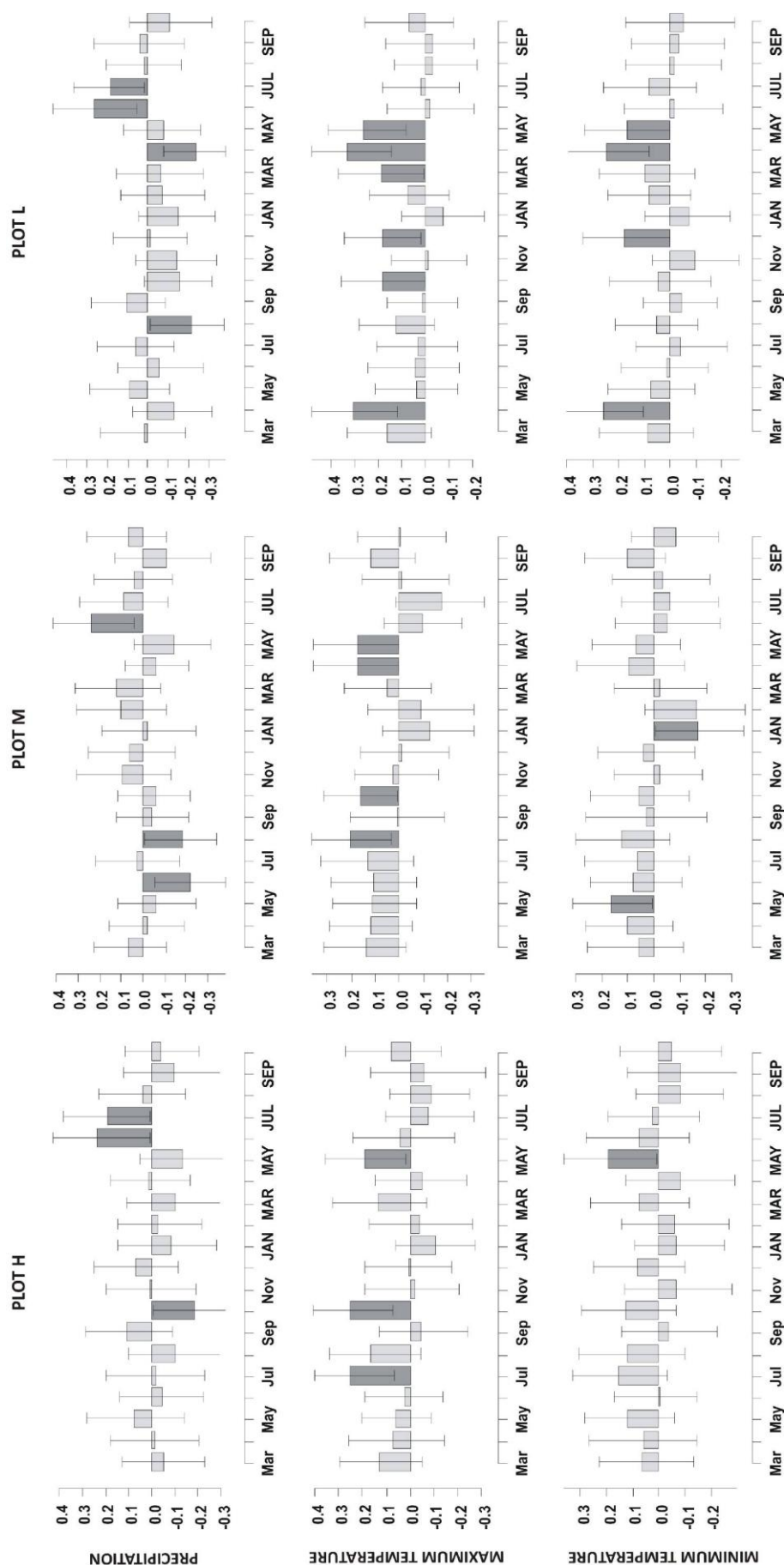


Fig. 3.3 Bootstrap correlation values between mean tree-ring indexed chronologies of the three altitudinal plots and total monthly precipitation, mean maximum and minimum monthly temperature. Correlations were calculated separately for each month for the period from March of the previous year (lowercase letters) to October for the current year (uppercase letters). The darker bars indicate a coefficient significant at $P < 0.05$, the lines represent the 95%-confidence interval.

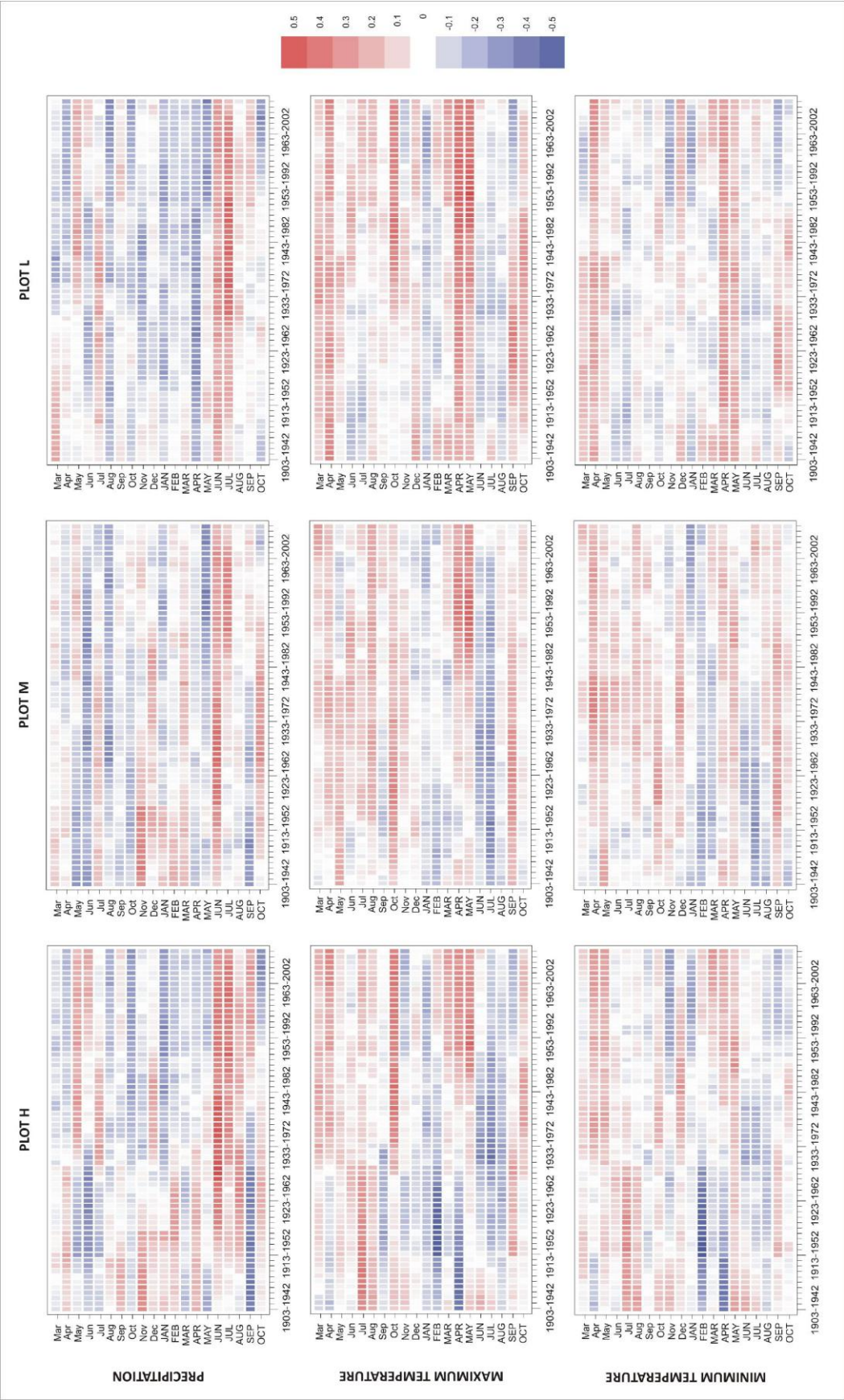


Fig. 3.4 The 40-year running means of bootstrapped statistically significant correlation coefficients between climate data and tree ring indexed chronologies for AD 1902–2008. Significant correlation coefficients ($P < 0.05$) are < -0.2 or > 0.2 . Y axis: Correlations were calculated separately for each month for the period from March of the previous year (lowercase letters) to October for the current year (uppercase letters). Each rectangle represents a correlation calculated over a 40-year period plotted at the last year of each 40-year period. Colour scale corresponds to both the sign and strength of the correlations.

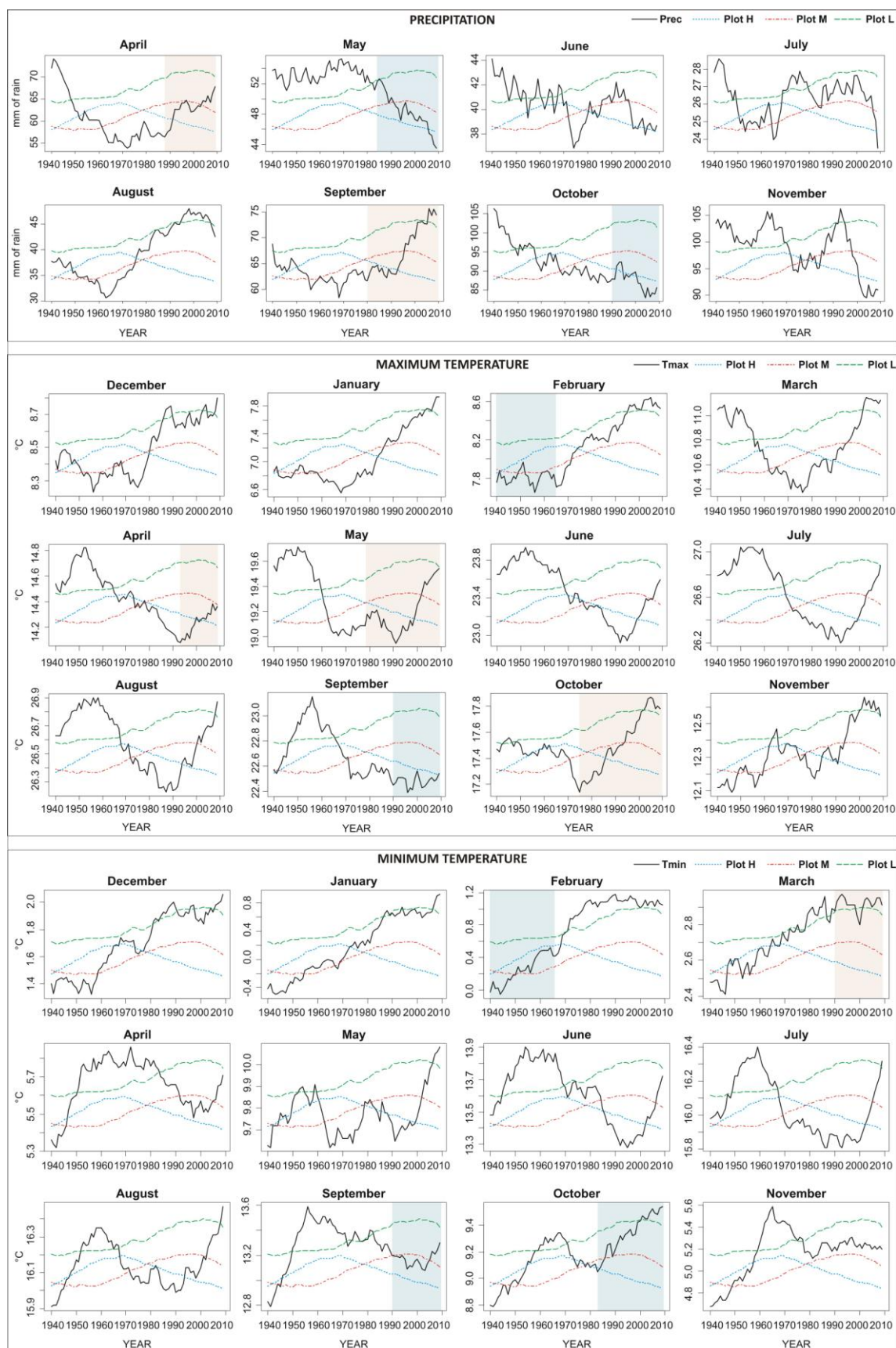


Fig. 3.5 Trend of raw tree-ring series and climatic variables using a 40-year moving window. Light-red and light-blue bars indicate, respectively, the positive or negative effect of climatic variables on tree-ring growth.

3.4 Discussion

In the Majella massif, climate variables affected mountain pine tree growth at the treeline ecotone, as demonstrated by significant correlations found with the spring maximum temperatures and summer precipitation. Our data suggest that, on the Majella massif, the tree line evolution is climatic-related. Several dendrochronological studies stressed the displacement of bioclimatic limits as the main growth-determining factors at the timberline for coniferous species on the Alps (e.g. Oberhuber 2004; Leonelli et al. 2009; Carrer et al. 2010), but only Piermattei et al. (2012) addressed these studies on the Apennines treeline. Nevertheless, only a few studies analysed mountain pine growth dynamics at the subalpine belt and, again, all of these were conducted in alpine environment (Dirnböck et al. 2008; Lingua et al. 2008), in particular for geomorphological aims (Santilli and Pelfini 2002; Pelfini et al. 2006). In the central Italian Alps, the climate of summer months has the strongest influence on mountain pine tree-ring growth, especially May and July mean temperature and June precipitation (Pelfini et al. 2006).

Our results, instead, would suggest a positive effect of spring maximum temperature, summer temperature being a minor climatic factor affecting mountain pine growth in the Majella massif, contrary to what happens for stone pine in the Alps (Carrer et al. 2007). In our site, it is likely that mild spring temperatures would be advantageous in expanding the vegetation period, through the full exploitation of the proximity to the sea and the irregular precipitation regimes of the Mediterranean-type climate. Low temperature and abundant snowfall during spring might delay bud burst, reducing growth time span, while warmer air would induce prompt shoot development, resulting in larger ring widths.

In Mediterranean climates, water is the most limiting factor to plant life, and a small change in site-specific water availability could have a great influence on stand composition, structure, and function (Joffre and Rambal 2002). Relatively high annual precipitation in the study area could support the positive correlation of radial growth of mountain pine with summer precipitation, particularly at the lower limit (Plot L), pointing to the Mediterranean mark of this forest ecosystem. However, the high solar radiation coupled with a relatively open canopy, particularly at the upper limit (Plot H), and the high soil permeability due to karst and fissuring of limestone could induce high evaporative water loss and low water holding capacity, explaining the general negative influence of summer temperature (Fig. 3.3). Lehner and Lütz (2002) found that mountain pine photosynthesis have high stability under changing climatic conditions, related to the better protection of twigs by snow cover because of their growth habits; though results of MCF showed that mountain pine is also a sensitive species, in terms of tree growth.

Essentially, three months played a key role in mountain pine growth: April, May and October. The first two represent the start of the growing season for mountain pine, while the last characterizes the end of it (Palombo 2013). At mid-lower altitude (Plot M and Plot L), the snow starts to melt earlier than at higher altitudes and precipitation would play a key role affecting negatively pine growth. This is probably because spring temperature would generally benefit the mountain pine and optimal precipitation could actually trigger the growing season or anticipate it. In fact, an increase in the amount of precipitation in April, during the years 1950-1990, was reflected in less negative correlation coefficients in Plot L (Fig. 3.4). At high altitude (Plot H), instead, mountain pine growth would depend also on the temperature, being the cause of snowmelt and then of water availability and soil warming. Considering that the vegetation season at higher altitude starts late and ends early, because of the temperature, the additional role of September precipitation might be considered important. The increased water availability by the end of the growing season, though coinciding with decreasing temperatures, would postpone the end of the tracheids maturation and allow a longer vegetation season, triggering next year's bud and needle development and hence favouring foliage expansion. Lehner and Lütz (2002) demonstrated that mountain pine have some advantages with high winter precipitation, since most needles would be covered by snow and thus protected from extreme temperatures. Nevertheless the direct influence of October precipitation of the current year on ring growth is difficult to explain, being October the month when the tracheids end the maturation phase (Palombo 2013).

Several conifers growing at arid and semi-arid sites correlate significantly with the climate preceding the growth period (Fritts 1971; Peterson and Peterson 2001; Littell et al. 2008). Douglas-fir growth, for instance, was reported to correlate significantly with late summer precipitation of the year prior to growth (Littell et al. 2008). Peterson and Peterson (2001) observed that mountain hemlock growth was also negatively correlated with previous year summer temperature, but positively with precipitation, since with larger cone production (suitable summer temperature) fewer resources will remain to support next year's radial growth. Yu et al. (2007) suggested that warmer October would be helpful to enhance the root activity of *Betula ermanii* Charm. at the treeline, conducing to the formation and survival of winter bud, which could promote a better growth in the next spring. A positive effect of temperatures of the previous October on pine growth throughout the whole altitudinal gradient, particularly in Plot H, was observed. High photosynthetic rates in the fall could lead to high carbohydrate storage and thus to increased growth in the following year (Fritts 1976). A delay in stopping further cell differentiation at the cambium level would suggest that optimal climatic condition in October might result in a longer growing season for mountain pine. A similar effect of previous autumn temperatures on subsequent year tree growth was reported by Oberhuber (2004)

and Carrer et al. (2007) for *Pinus cembra* and by Savva et al. (2006) for *Picea abies*, in the alpine timberline ecotone.

High mountain systems are considered to be thermally controlled (Beniston 2000, 2003; Körner and Paulsen 2004), and warming temperatures should result in their upward expansion (Moen 2006). However, land use in subalpine and alpine areas (grazing and logging) affects the flora distribution just as much as climate (Švajda et al. 2011). In Mediterranean mountain systems, direct and indirect effects of land use changes are particularly significant due to long-standing human impact on the original landscape heterogeneity, and to the intense changes in structural and functional features for the majority of forests (Marchetti et al. 2010). Our results would confirm the climatic effects on mountain pine growth. The tree line position (the uppermost or northernmost limit of tree growth form) represents a well-known climatic boundary (Compostella et al. 2012), though at the same time, being strongly influenced by human activity. As in other European mountain systems (e.g. Dirnböck et al. 2003; Améztegui et al. 2010; Švajda et al. 2011; Mihai et al. 2012; Piermattei 2012), also the subalpine and alpine belts of the Majella massif, during the last 50 years, have been influenced by land abandonment, grazing cessation, and switch from traditional timber use (construction and firewood) to more recent materials (Palombo et al. in press). This has been particularly evident following 1995, when the MNP was established, enabling the secondary forest succession, which has led to the colonization of previously abandoned areas and to an upward shift of *Pinus mugo* dwarf-shrubland (Stanisci 1997; Di Giustino et al. 2002; van Gils et al. 2008; Palombo et al. in press).

Several studies have demonstrated the upward expansion of mountain pine at European level (e.g. Dirnböck et al. 2003; Gehrig-Fasel et al. 2007; Améztegui et al. 2010; Švajda et al. 2011; Mihai et al. 2012), a few also describing the loss of surface of mountain pine krummholz because of the upward expansion of timberline forests; this was the case of spruce forests in the Iezer Mountains (Mihai et al. 2012) or in Slovakia (Mind'as et al. 2004). We hypothesize that the recent upward expansion of beech forest (van Gils et al. 2008) on the Majella massif might indirectly drive the shift of mountain pine ecotone upward. Despite a climatic change that would be adverse (drier) and the proliferation of large forest ungulates intensively browsing seedlings and saplings, the present-day expansion of beech forest suggests that it is mostly a result of land-use abandonment and decrease of impact of human practices on forests of the MNP. Nevertheless, these trees do not necessarily respond uniformly to climatic changes, tree-climate responses depending on the individuals' position within the mountain pine-treeline ecotone, as well as on other non-climatic factors, such as intra- and inter-specific competition, other than disturbances.

3.5 Conclusions

Results show that mountain pine is very sensitive to climate, particularly to the temperature at the beginning and at the end of the growing season and to summer precipitation. This shows that mountain pine at this site forms a treeline strongly influenced by climate. This tree line is not only limited by summer temperature, as usual for Alpine treeline (Oberhuber 2004; Pelfini et al. 2006; Carrer et al. 2007) but also by summer precipitation, which is peculiar for such Mediterranean mountain ecosystem (Camarero et al. 2010).

This study revealed the dendrochronological potential of mountain pine, which was often considered as a difficult species to be studied with a dendrochronological approach, because of the narrow and the high variable ring growth, related to the prostrate habits of mountain pine. In our case, we used mountain pine chronologies to demonstrate the climatic sensitivity of this species and to analyse the variation of mountain pine response to climate change. Growing above the limit of tree vegetation, mountain pine proved to be a promising species for monitoring climate- and land use-driven influences over tree ring growth at high elevation. Even if we did not find a sharp variation in tree ring responses to climate change, we could confirm the possibility of a lengthening of the growing season, according to the current climate trends and future scenarios. Although transects across a tree line might not reveal spatial changes from forest interior to forest edge in Mediterranean mountains, where tree line has shifted repeatedly, tree-line studies will likely reveal evidence of both historical legacies and ecological succession. How trees optimize the trade-offs between tree-ring development and foliage expansion warrants further studies, considering ecophysiological responses to environmental conditions.

Despite the relatively high annual precipitation in the study area, in comparison with lower elevation, the radial growth of mountain pine was found to be highly correlated with precipitation. This result may be partly explained by soil conditions found on our study sites, which are characterized by thin soil depth, high content of limestone, and low water holding capacity. Because mountain pine is also affected by moisture, radial growth (and needle development) at the tree line in the MNP could not be predicted by temperature alone even under a scenario of global warming. Mountain pine expansion will certainly have an impact on the distribution of endemic-rich arctic-alpine vegetation on the Majella massif, because of the climate and land use changes, causing great habitat fragmentation and high biodiversity loss. These aspects are particularly important for the MNP authorities, which are called to plan the appropriate management of forest resources towards the preservation of biodiversity.

Acknowledgements We acknowledge Giovanni Santopuoli, Mauro Maesano, Lorenzo Sallustio, and Claudia Coccozza for field assistance. We are grateful to Patrick Fonti, Hein van Gils, Elena Liberatoscioli, Giovanni Pelino, and Edoardo Micati for valuable suggestion and fruitful discussion. This work was possible thanks to the authorities of the MNP that allowed the sampling in the study areas.

Funding Part of the financial support for Caterina Palombo was provided by a MNP fellowship.

References

- Améztegui A., Brotons L., Coll L. 2010. Land-use changes as major drivers of mountain pine (*Pinus uncinata* Ram.) expansion in the Pyrenees. *Global Ecology and Biogeography* 19: 632-641.
- Beniston M. 2000. Environmental Change in Mountains and Uplands. London, United Kingdom: Arnold.
- Beniston M. 2003. Climatic change in mountain regions: a review of possible impacts. *Climatic Change* 59: 5–31.
- Blasi C., Di Pietro R., Fortini P., Catonica C. 2003. The main plant community types of the alpine belt of the Apennine chain. *Plant Biosystems* 137: 83-110.
- Blasi C., Di Pietro R., Pelino G. 2005. The vegetation of alpine belt karst-tectonic basins in the Central Apennines. *Plant Biosystems* 139: 357-385.
- Boden S., Pyttel P., Eastaugh C.S. 2010. Impacts of climate change on the establishment, distribution, growth and mortality of Swiss stone pine (*Pinus cembra* L.). *iForest* 3: 82-85.
- Briffa K.R. 1995. Statistical aspects of the interpretation of high-resolution proxy climate data: the example of dendroclimatology. In *Analysis of climate variability: applications of statistical techniques*, Von Storch, H. and Navarra, A. (eds.). Springer, Berlin, Germany, pp. 77-94.
- Camarero J.J., Gutiérrez E., Fortin M.J. 2006. Spatial patterns of plant richness across treeline ecotones in the Pyrenees reveal different locations for richness and tree cover boundaries. *Global Ecology and Biogeography* 15: 182-191.
- Camarero J.J., Olano J.M., Parras A. 2010. Plastic bimodal xylogenesis in conifers from continental Mediterranean climates. *The New phytologist* 185: 471–80.
- Carrer M., Nola P., Eduard J.L., Motta R., Urbinati C. 2007. Regional variability of climate-growth relationships in *Pinus cembra* high elevation forests in the Alps. *Journal of Ecology* 95: 1072–1083.
- Carrer M., Nola P., Motta R., Urbinati C. 2010. Contrasting tree-ring growth to climate responses of *Abies alba* toward the southern limit of its distribution area. *Oikos* 119: 1515–1525.

- Compostella C., Trombino L., Caccianiga M. 2012. Late Holocene soil evolution and treeline fluctuations in the Northern Apennines. *Quaternary International*. ISSN 1040-6182, 10.1016/j.quaint.2012.02.011.
- Cook E.R. and Peters K. 1981. The smoothing spline: a new approach to standardizing forest interior tree-ring width series for dendroclimatic studies. *Tree-Ring Bulletin* 41: 45–53.
- Cook E.R. and Holmes R.L. 1984. Program ARSTAN users manual. Laboratory of Tree Ring Research, University of Arizona.
- Di Giustino A., Stanisci A., Acosta A., Blasi C. 2002. Il limite superiore della faggeta nella Majella occidentale (Abruzzo). *Informatore Botanico Italiano* 34: 71–78.
- Dirnböck T., Dullinger S., Grabherr G. 2003. A regional impact assessment of climate and land-use change on alpine vegetation. *Journal of Biogeography* 30: 401–417.
- Dirnböck T., Dullinger S., Köck R. 2008. Organic matter accumulation following *Pinus mugo* Turra establishment in subalpine pastures. *Plant Ecology and Diversity* 1: 59–66.
- Fritts H.C. 1971. Dendroclimatology and Dendroecology. *Quaternary Research* 1: 419–49.
- Fritts H.C. 1976. Tree-rings and climate. Academic Press, London.
- Gehrig-Fasel J., Guisan A., Zimmermann N.E. 2007. Treeline shifts in the Swiss Alps: Climate change or land abandonment? *Journal of Vegetation Science* 18: 571–582.
- Giraudi C. 2003. Middle to late Holocene glacial variations, periglacial processes and alluvial sedimentation on the higher Apennine massifs (Italy). *Quaternary Research* 64: 176–184.
- Gottfried M., Pauli H., Futschik A., Akhalkatsi M., Barančok P., Benito Alonso J.L., Coldea G., Dick J., Erschbamer B., Fernández Calzado M.R., Kazakis G., Krajči J., Larsson P., Mallaun M., Michelsen O., Moiseev D., Moiseev P., Molau U., Merzouki A., Nagy L., Nakhutsrishvili G., Pedersen B., Pelino G., Puscas M., Rossi G., Stanisci A., Theurillat J.P., Tomaselli M., Villar L., Vittoz P., Vogiatzakis I., Grabherr G. 2012. Continent-wide response of mountain vegetation to climate change. *Nature Climate Change* 2: 111–115.
- Grace J., Berninger F., Nagy L. 2002. Impacts of Climate Change on the Tree Line. *Annals of Botany* 90: 537–544.
- Guiot J. 1991. The bootstrapped response function. *Tree-Ring Bulletin* 51: 39–41.
- Hamilton L.S., Gilmour D.A., Cassels D.S. 1999. Forêts et silviculture en montagne. In: Messerli B, Ives JD. *Les montagnes dans le monde. Une priorité pour un développement durable*, pp. 249–278. Glénat, Grenoble, France.
- Holmes R.L. 1983. Computer assisted quality control in tree-ring dating and measurement. *Tree-Ring Bulletin* 43: 69–78.

- Holtmeier F.K. 1981. What does the term 'krummholz' really mean? Observations with special reference to the Alps and the Colorado Front Range. *Mountain Research and Development* 1: 253-260.
- Jodłowski M. 2006. Geographical controls on the course of the upper mountain pine (*Pinus mugo*) limit in the Tatra mts. *Ekologia* 25: 105-114.
- Joffe R. and Rambal S. 2002. Mediterranean ecosystems. Encyclopedia of Life Sciences. Macmillan Publishers Ltd., London.
- Körner C. and Paulsen J. 2004. A world-wide study of high altitude treeline temperatures. *Journal of Biogeography* 31: 713-732.
- Lehner G. and Lütz C. 2003. Photosynthetic functions of cembran pines and dwarf pines during winter at timberline as regulated by different temperatures, snowcover and light. *Journal of plant physiology* 160: 153-66.
- Lenoir J., Gégout J.-C., Marquet P.A., de Ruffray P., Brisse H. 2008. A significant upward shift in plant species optimum elevation during the 20th century. *Science* 320: 1768-1771.
- Leonelli G., Pelfini M., Battipaglia G., Cherubini P. 2009. Site-aspect influence on climate sensitivity over time of a high-altitude *Pinus cembra* tree-ring network. *Climatic Change* 96: 185-201.
- Linares J.C., Camarero J.J., Carreira J.A. 2009. Interacting effects of changes in climate and forest cover on mortality and growth of the southernmost European fir forests. *Global Ecology and Biogeography* 18: 485-497.
- Lingua E., Cherubini P., Motta R., Nola P. 2008. Spatial structure along an altitudinal gradient in the Italian central Alps suggests competition and facilitation among coniferous species. *Journal of Vegetation Science* 19: 425-436.
- Littell J.S., Peterson D.L., Tjoelker M. 2008. Douglas-fir growth in mountain ecosystems: water limits tree growth from stand to region. *Ecological Monographs* 78: 349-68.
- MacDonald D., Crabtree J.R., Wiesinger G., Dax T., Stamou N., Fleury P., Gutierrez L.J., Gibon, A. 2000. Agricultural abandonment in mountain areas of Europe: Environmental consequences and policy response. *Journal of Environmental Management* 59: 47-69.
- Marchetti M., Tognetti R., Lombardi F., Chiavetta U., Palumbo G., Sellitto M., Colombo C., Iovieno P., Alfani A., Baldantoni D., Barbatì A., Ferrari B., Bonacquisti S., Capotorti G., Copiz, R., Blasi, C. 2010. Ecological portrayal of old-growth forests and persistent woodlands in the Cilento and Vallo di Diano National Park (southern Italy). *Plant Biosystems* 144: 130-147.
- Menzel A., Sparks T.H., Estrella N., Koch E., Aasa A., Ahas R., Alm-Kübler K., Bissolli P., Braslavská O., Briede A., Chmielewski F.M., Crepinsek Z., Curnell Y., Dahl, Å., Defila C., Donnelly A., Filella Y., Jatczak K., Måge F., Mestre A., Nordli Ø., Peñuelas J., Pirinen P., Remišová V., Scheffinger H., Striz M., Susnik A., van Vliet A.J.H., Wielgolaski F.-E., Zach S., Züst, A. 2006. European

- phenological response to climate change matches the warming pattern. *Global Change Biology* 12:1969–1976.
- Migliaccio F. 1966. La vegetazione a *Pinus pumilio* della Majella. *Annali di Botanica* (Roma) 28: 539–550.
- Mihai B., Savulescu I., Sandric I. 2007. Change detection analysis (1986–2002) of vegetation cover in Romania. *Mountain Research and Development* 27: 250–258.
- Mind'áš J., Čaboun V., Priwitzer T. 2004. Timber line and expected climate changes [in Slovak]. Kadlečík J. (ed.). Year-book Turiec and Fatra. ŠOP SR, Vrútky, Slovakia, pp. 17–23.
- Mitchell T.D. and Jones P.D. 2005. An improved method of constructing a database of monthly climate observations and associated high-resolution grids. *International Journal of Climatology* 25: 693–712.
- Moen J. 2006. Treeline dynamics in a changing climate. In *Global Change in Mountain Regions*, Price M. (ed.). Sapiens, London, United Kingdom, pp. 187–188.
- Nicolini G., Viola F., Zucca M., Chemini C. 2002. The Italian mountains. *Unasyva* 53: 68–70.
- Nicolussi K. and Patzelt G. 2006. Klimawandel und Veränderungen an der alpinen Waldgrenze—Aktuelle Entwicklungen im Vergleich zur Nacheiszeit. *BFW-Praxisinformation* 10: 3–5.
- Oberhuber W. 2004. Influence of climate on radial growth of *Pinus cembra* within the alpine timberline ecotone. *Tree physiology* 24: 291–301.
- Palombo C. 2013. The influence of land-use and climatic changes on mountain pine (*Pinus mugo* Turra spp. *mugo*) ecotone dynamics at its southern range margin on the Majella massif, Central Apennines. Ph.D. thesis, University of Molise, Campobasso, Italy.
- Palombo C., Chirici G., Marchetti M., Tognetti R. 2013. Is land abandonment affecting forest dynamics at high elevation in Mediterranean mountains more than climate change? *Plant biosystems* in press.
- Pelfini M., Leonelli G., Santilli M. 2006. Climatic and environmental influences on mountain pine (*Pinus montana* Miller) growth in the central Italian Alps. *Arctic, Antarctic, and Alpine Research* 38: 614–623.
- Peñuelas J., Hunt J.M., Ogaya R., Jump A.S. 2008. Twentieth century changes of tree-ring $\delta^{13}\text{C}$ at the southern range-edge of *Fagus sylvatica*: increasing water-use efficiency does not avoid the growth decline induced by warming at low altitudes. *Global Change Biology* 14: 1076–1088.
- Peterson D.W. and Peterson D.L. 2001. Mountain hemlock growth responds to climatic variability at annual and decadal time scales. *Ecology* 82: 3330–45.
- Piermattei A., Renzaglia F., Urbinati C. 2012. Recent expansion of *Pinus nigra* Arn. above the timberline in the central Apennines, Italy. *Annals of Forest Science* 69: 509–517.

- Randin C.F., Engler R., Normand S., Zappa M., Zimmermann N.E., Pearman P.B., Vittoz P., Thuiller W., Guisan A. 2009. Climate change and plant distribution: local models predict high-elevation persistence. *Global Change Biology* 15: 1557–1569.
- Rolland C., Petitcolas V., Michalet, R. 1998. Changes in radial tree growth for *Picea abies*, *Larix decidua*, *Pinus cembra* and *Pinus uncinata* near the alpine timberline since 1750. *Trees* 13: 40-53.
- Santilli M. and Pelfini M. 2002. Dendrogeomorphology and dating of debris flows in the Valle del Gallo, Central Alps, Italy. *Dendrochronologia* 20: 269–284.
- Savva Y., Oleksyn J., Reich P.B., Tjoelker M.G., Vaganov E.A., Modrzynski, J. 2006. Interannual growth response of Norway spruce to climate along an altitudinal gradient in the Tatra Mountains, Poland. *Trees* 20: 735–746.
- Saxe H., Cannell M.G.R., Johnsen B., Ryan M.G., Vourlitis G. 2001. Tree and forest functioning in response to global warming. *New Phytologist* 149: 369–399.
- Schueller J.F. and Rolland C. 1995. Influence de l'altitude, de l'exposition et du climat sur la croissance du pin à crochets (*Pinus uncinata* Ram.) en Cerdagne (Pyrénées Orientales Françaises). *Pirineos* 145–146: 23–34.
- Schweingruber F.H. 1988. Tree rings: basics and applications of dendrochronology. Kluwer Academic Press, Dordrecht.
- Stanisci A. 1997. Gli arbusteti altomontani dell'Appennino centrale e meridionale. *Fitosociologia* 34: 3-46.
- Stanisci A., Lavieri D., Acosta A., Blasi C. 2000. Structure and diversity trends at *Fagus* timberline in central Italy. *Community Ecology* 1: 133–138.
- Stanisci A., Pelino G., Blasi C. 2005. Vascular plant diversity and global change in central Apennine (Italy). *Biodiversity and Conservation* 14: 1301-1318.
- Stanisci A., Carranza M.L., Pelino G., Chiarucci A. 2010. Assessing the diversity pattern of cryophilous plant species in high elevation habitats. *Plant Ecology* 212: 595–600.
- Švajda J., Solár J., Janiga M., Buliak, M. 2011. Dwarf pine (*Pinus mugo*) and selected abiotic habitat conditions in the western Tatra Mountains. *Mountain Research and Development* 31: 220-228.
- Thomas C.D., Cameron A., Green R.E., Bakkenes M., Beaumont L.J., Collingham Y.C., Erasmus B.F.N., De Siqueira M.F., Grainger A., Hannah L., Hughes L., Huntley B., Van Jaarsveld A.S., Midgley G.F., Miles L., Ortega-Huerta M.A., Peterson A.T., Phillips O.L., Williams, S.E. 2004. Extinction risk from climate change. *Nature* 427: 145–8.
- Thuiller W., Albert C., Araújo M.B., Berry P.M., Cabeza M., Guisan A., Hickler T., Midgley G.F., Paterson J., Schurr F.M., Sykes M.T., Zimmermann N.E. 2008. Predicting global change

- impacts on plant species' distributions: Future challenges. *Perspectives in Plant Ecology, Evolution and Systematics* 9: 137–152.
- van Gils H., Batsukh O., Rossiter D., Munthali W., Liberatoscioli E. 2008. Forecasting the pattern and pace of *Fagus* forest expansion in Majella National Park, Italy. *Applied Vegetation Science* 11: 539–546.
- van Gils H., Odoi J.O., Andrisano T. 2010. From monospecific to mixed forest after fire? An early forecast for the montane belt of Majella, Italy. *Forest Ecology and Management* 259: 433–439.
- van Gils H., Conti F., Ciaschetti G., Westinga E. 2012. Fine resolution distribution modelling of endemics in Majella National Park, Central Italy. *Plant Biosystems* 146: 276–287.
- Watson C. 1996. The vegetational history of the northern Apennines, Italy: information from three new sequences and a review of Regional vegetational change. *Journal of Biogeography* 23: 805–841.
- Way D.A. 2011. Tree phenology responses to warming: spring forward, fall back? *Tree physiology* 31: 469–71.
- Whitehead F. 1951. Ecology of the altipiano of Monte Maiella, Italy. *Journal of Ecology* 39: 330–355.
- Wigley T.M.L., Briffa K.R., Jones P.D. 1984. On the average value of correlated time series with application in dendroclimatology and hydrometeorology. *Journal of Climate and Applied Meteorology* 23: 201–221.
- Yu D., Wang G.G., Dai L., Wang Q. 2007. Dendroclimatic analysis of *Betula ermanii* forests at their upper limit of distribution in Changbai Mountain, Northeast China. *Forest Ecology and Management* 240: 105–113.
- Zang C. and Biondi F. 2012 Dendroclimatic calibration in R: The bootRes package for response and correlation function analysis. *Dendrochronologia* doi:10.1016/j.dendro.2012.08.001.

FOURTH CHAPTER:

Timing of compression and opposite wood formation in mountain pine (*Pinus mugo* Turra ssp. *mugo*) on the Majella massif, Southern Apennines, Italy.

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ABSTRACT

At the alpine tree line, low temperature limits cambial activity and determines secondary growth. The ring width is in fact a commonly used proxy for growing season temperatures. However, large differences in width exist, if rings include reaction wood. This study aims at investigating if these differences result from differing timing of wood formation, being expected to be longer in the reaction wood.

As study object we considered mature and partially prostrating mountain pines growing above the timberline (1900 m a.s.l.) on the Majella massif, Italy. The sampling has been performed weekly from May to October 2011 on 10 trees by micro-coring the compression wood (CW) and the opposite wood (OW) where the branch curvature was stronger.

Results indicated that CW growth ring had around 1.5 times more cells than OW and that there were differences in the duration, timing and amount of cells in the different phenological phases. Although the onset of the first enlargement initiated simultaneously in both types of wood (on 20 May), cell differentiation completed two week earlier in OW (on 22 September) than in CW (on 6 October). Moreover, we observed that while for the division and enlargement phases there were no differing cell number, for the cell wall thickening there were 2.5 times more cells in CW than in OW.

Based on this study, it appears that wood formation occurs at faster rate in CW than in OW. These differences result from a longer time for lignifying the more numerous tracheids.

4.1 Introduction

The period in which wood formation occurs is the time window when xylem is differentiating and environmental factors can act directly on the cells constituting the tree ring (Frankenstein et al. 2005). Wood-anatomical variability along tree-ring sequences could be used to elucidate how individual trees and species respond to changing environmental conditions occurring during the growing season (Schweingruber 1996, 2006; Fonti et al. 2010). There are cases, however, in which disturbances are not merely concentrated during the growing season, as for example mechanical stress due to snow pressure along steep slopes during winter season. In these cases, woody plants can react to such stresses only during the growing season, by modifying their xylem structure and growth. This phenomenon happens every year to dwarf mountain pine (*Pinus mugo* spp. *mugo*) growing at the treeline ecotone on the Majella massif (Majella National Park – MNP, Italy), where it forms the «krummholz mat» (Lehner and Lütz 2003). During winter, wood fibres must hold the weight of the snow on the canopy; although, as it accumulates, it may start breaking the branches. During spring, twigs need to regain their vertical orientation by creating expansion forces on the compression side of the stem. Mountain pine can survive in such particular environment because it

has a very flexible stem that re-establish a more or less vertical position after snow melt. This important feature is strictly connected to a special tissues forming the stems and twigs, i.e. the compression wood (CW). In most gymnosperms, this tissue is formed on the lower side of branches and leaning stems, exerting an active pressure along the longitudinal direction generating movements in the desired direction (Timell 1973). In comparison with normal wood (NW), CW is characterized by: greater amounts of lignin and galactan and smaller amounts of cellulose and galactoglucomannan (Timell 1986; Donaldson 2001; Nanayakkara et al. 2005, 2009); absence of a tertiary cell wall (S3) layer; most thickening and enhanced lignification of the outer secondary wall, designed as S2_L layer (Côté et al. 1968); presence of helical cavities and higher microfibril angle in the secondary cell wall (S2) (Donaldson et al. 2004); tracheids with a more rounded appearance in cross section; presence of intracellular spaces and shorter tracheids (Timell 1986). In CW, the cell wall of tracheid show reduced lignification of the middle lamella (M) and increased lignification of the S2_L layer (Côté et al 1968; Singh and Donaldson 1999; Donaldson et al. 1999; Donaldson 2001). However, it is only the so-called typical CW tracheid that shows all these structural features; while, intermediate degrees of development occur in a continuum from NW or opposite wood (OW) to mild and severe CW. It results that, in comparison to the NW, the compression wood is characterized by higher specific gravity, reduced permeability, lower radial and tangential shrinkage, substantially larger longitudinal shrinkage, higher compressive strength, lower tensile strength, lower modulus of elasticity and is more brittle (Donaldson and Turner 2001; Gardiner and Macdonald 2005). These properties, although causing major problems in the utilisation of sawn timber, make CW an excellent material from a functional aspect in living plants (Gardiner and Macdonald 2005).

The formation of compression wood, since usually characterized by more dense and larger rings, requires more resources in terms of carbohydrates and timing of formation. Considering that the growth rings in CW are generally wider than those in OW (Timell 1973), it is still unknown how the differentiation of anatomical characteristics in the same tree can act simultaneously but differently for opposite growth directions. Several studies investigated the dynamics of wood formation in different species or trees for comparing the effects of natural events, including elevation, temperature or age (Rossi et al. 2008a, 2008b; Moser et al. 2010; Prislan et al. 2011), or induced by treatments, as drought, heating and cooling (Gričar et al. 2006, 2007; Eilman et al. 2009; Galle et al. 2010; Fonti et al. 2013), on the initiation, termination, rate and extent of growth. Only one study (Anfodillo et al. 2012) described the sequence of the phenological phases along the same stem. Lupi et al. (2010) stated that the onset of xylem differentiation drives all processes of tree-ring formation by triggering the successive timings of cell production and maturation. If this assumption is true we

would expect that the formation of compression wood either need a longer vegetation season or occurs at a faster rate than the normal wood.

In this study, for the first time, we analysed timing of CW and OW formation in mountain pine, throughout the growing season in order to identify how, in term of xylogenesis, the shrub realize the production of CW. To achieve this goal we assessed intra-annual dynamics of cambial activity in both CW and OW in mountain pine, during the 2011 growing season.

4.2 Materials and methods

4.2.1 Study site

The study site ($42^{\circ} 8' 50''$ N, $14^{\circ} 6' 18''$ E - Fig. 4.1) is located at the subalpine-alpine ecotone of the State Natural Reserve “Valle dell’Orfento”, within the MNP. An area of about 1.5 ha were established at 1900 m a.s.l., at the southern limit of the *krummholz mat* distribution, where the contiguous beech forest gives way to the mountain pine woodlands.

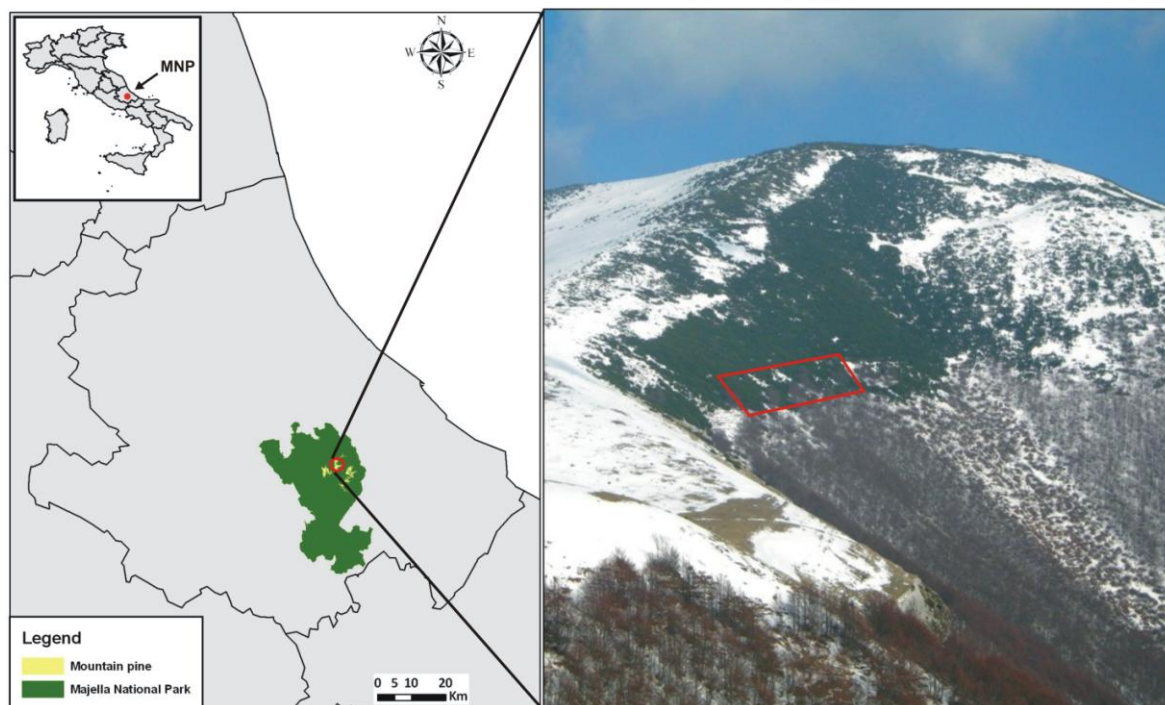


Fig. 4.1 Location of the study area in the Majella National Park, Abruzzo region – Italy. It is clear from the picture (taken on 19 April) the timberline formed by European beech (*Fagus sylvatica* L.) and the subalpine ecotone dominated by mountain pine (*Pinus mugo* Turra spp. *mugo*).

The nearest meteorological station (Sant’Eufemia a Majella, 888 m a.s.l. - Servizio Idrografico) recorded for the period 1932-2009 a mean annual temperature of 10.7°C and annual total

precipitation of 1430 mm, mainly concentrated in autumn and winter. During the entire 2011 growing season, temperatures were recorded hourly by two data loggers (Omega Engineering). One was installed in the study site, at 2.5 m above the ground and protected from direct solar radiation, and the other was fixed inside a mountain pine stem, below the bark. Monitoring of temperature trend (Fig. 4.2) was performed in order to evaluate possible effects of temperature on xylem formation.

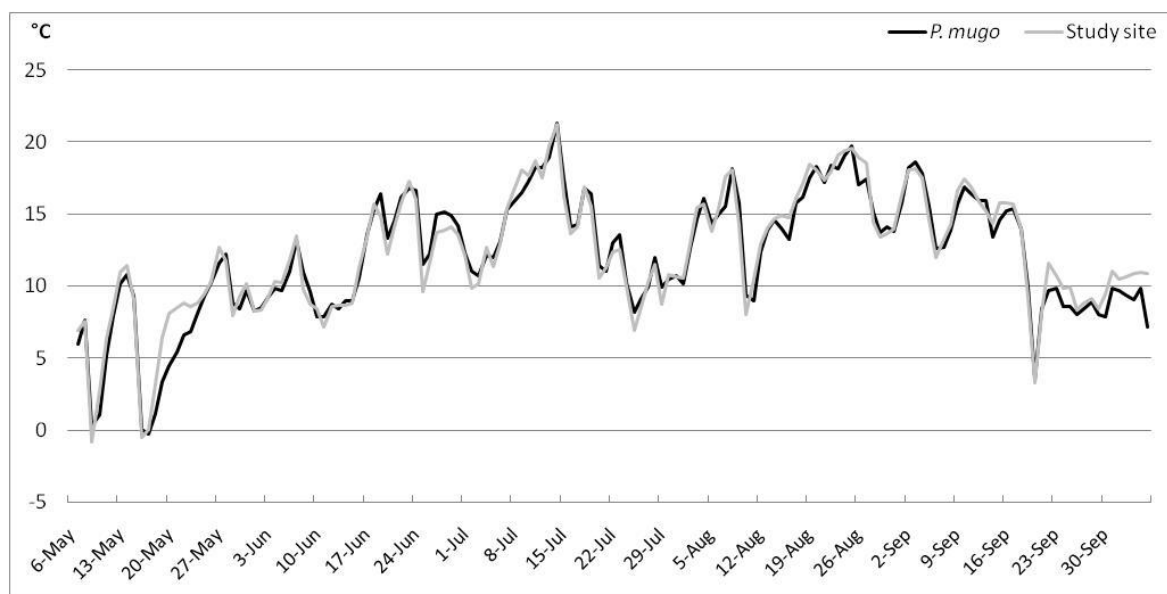


Fig. 4.2 Trends of mean daily temperature recorded from May to October 2011. The grey line was obtained from the study site (2.5 m above the ground), the black one from the mountain pine stem

4.2.2 Monitoring xylem formation

The sampling was performed weekly, from 6 May to 6 October 2011. Samples of tissues containing the bark, cambium and the last formed tree-ring were collected from mountain pines by micro-coring, through the Trephor tool (Rossi et al. 2006a), both the CW and the OW, where the curvature of the stem was accentuated. Every week, 10 different plants were randomly sampled, in order to avoid wound effects caused by previous sampling, because of the small size of mountain pine stem. A total of 420 microcores (1.8 mm in diameter, 15 mm in length) were extracted. Immediately after the sampling, to preserve forming cells from degradation, the samples were put in Eppendorf vials with 75% ethanol and 25% acetic acid for 24 h and then stored in 70% alcohol solution (Moser et al. 2010). The samples were then prepared for cellular analysis following the procedure described in Schweingruber et al. (2006).

Transversal microsections about 10-12- μ m-thick were obtained using a sliding microtome. The microsections were then stained with Safranin and Astrablue, dehydrated with graded series of

ethanol and fixed to microscope slides with Canada balsam. Ring formation was assessed by anatomical observations with an Olympus BX 40 light microscope at a magnification of 400x. Observations consisted in counting the number of cells in the cambial zone (CZ) and tracheids in enlargement (E), wall thickening (WT) and mature (M) phases along three radial files on each microsection (Rossi et al. 2006b), in both CW and OW for comparison purposes. Flattened cells are typical of the cambial zone; an increase in the number of cells in the cambial zone indicates the start of its divisional activity. Enlarging cells are characterized by thin primary cell walls, with fluctuating radial diameter, roughly twice or more times that of dividing cambial cells. Cells in wall thickening, which present the maximum radial diameter, were easily discriminated by polarized light because of the birefringence of the secondary cell wall. The appearance of red colour in the secondary cell wall, stained by Safranin, indicated the start of its lignification. The end of wall thickening corresponds to the complete lignification of xylem cells; fully mature tracheids were recognized by red-stained cell walls and empty lumina. Average values of three radial directions were used for subsequent analysis (Gricar et al. 2006; Moser et al. 2010). Severity of CW of all the growing rings included in the microcores was assessed by the Yumoto et al. (1983) classification (Table 4.1, Fig. 4.3).

Moreover, the number of cells of the last formed ring (year 2010) was counted and used successively for standardizing the number of cells formed during the year 2011. This standardization procedure was necessary because of the great variability in ring width among the sampled trees. Standardized data were multiplied by 100 (to reach integer values > 1) in order to be used in the R (R Development Core Team, 2007) package CAVIAR (Rathgeber 2011; 2012) for:

- assessing critical dates and duration of CW and OW formation;
- assessing the dynamics of tracheids production in both CW and OW, and fitting the total number of cells by the Gompertz function (Rossi et al. 2003, Rathgeber 2011);
- comparing the duration of the different phenological phases between CW and OW.

Table 4.1 Grading system to classify different degrees of compression wood severity, according to Yumoto et al. (1983) revised from Timell (1986), based on the three features visible at the magnification of 400x.

Grade	Wall Thickness	Intercellular spaces (IS)	Boundary S1-S2 _t
S	Very thick	Present	Nearly circular
I	Thick	Present	Round but not circular
D	Relatively thick	Present or Absent	Round but variable depending on the pres./abs. of IS
C	Thicker than normal	Usually absent	Fairly round
B	Slightly thicker (similar or somewhat thinner than normal)	Generally absent	Slightly round
A	Similar to normal wood	Generally absent	Very faintly round

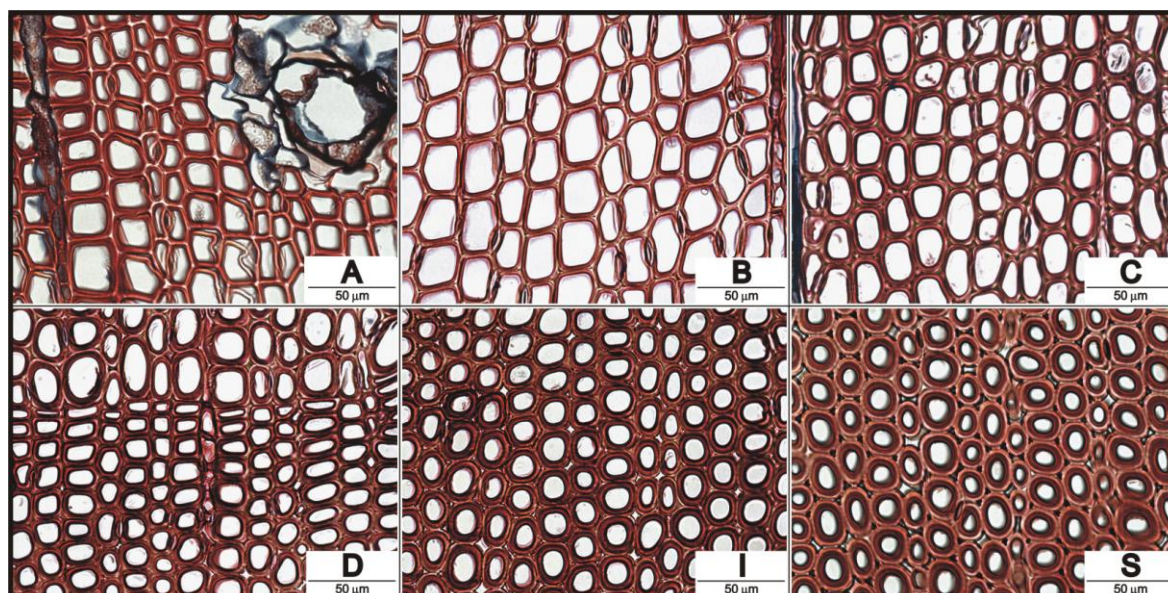


Fig. 4.3 Examples of different degree of compression wood severity, classified on the basis of the grading system described in Table 1.

4.3 Results

4.3.1 Wood structure

The first important result that emerges by counting the number of cells is the very high variability of radial growth between mountain pine individuals. Performing the sampling in 10 mountain pine individuals, chosen randomly every week, we observed that the number of cells forming growing rings in 2010 ranged from a maximum of 200 to a minimum of 14 tracheids in CW and from 133 to 4 tracheids in OW.

In addition, more numerous cells were found in CW rather than in OW (Fig. 4.5b). Out of 210 samples, the 74% of growing rings formed in 2010 presented a number of cells higher in CW than in OW, the 37% of which above 1.5 times and the 17% more than twice (Table 4.2).

4.3.2 Timings of wood production

The dormant cambium in mountain pine was composed of 4 to 7 closely spaced fusiform cells (Fig. 4.5d). At the first sampling date, on 6 May (day of the year, DOY 126), the cambial activity was already started in several samples, which presented 9 to 12 cells in the CZ, irrespective of the type of wood. The same happened during the second sampling, on 12 May (DOY 132), probably indicating the strong variability between individuals. On 20 May (DOY 140) cambial activity was already started in both types of wood; the number of dividing cambial cells increased from that time until the middle

of June, and then gradually decreased until the number of dormant cambium. During the growing season (May-October) the mean number of cambial cells was 12-13 in CW and OW, reaching the maximum value of 18-20 cells in CW (Fig. 4.4). At the same date (DOY 140), the first enlarging cells were observed. The maximum production of cells occurred around the middle of June (Fig. 4.5b), followed by a decrease in the number of enlarging cells (Fig. 4.4).

Table 4.2 Mean number of cells and standard deviation of the growing ring of 2010 in CW and OW. The type of CW of each microcore was assessed. *In brackets the number of microcores out of ten having the same type of CW indicated by the preceding letter.

2010-Tree Ring	OPPOSIT WOOD (OW)		COMPRESSION WOOD (CW)		
DATE	Mean n. cells	St.D. n. cells	Mean n. cells	St.D. n. cells	Type of CW (*)
06/05/2011	44.4	±17.1	63.6	±23.5	D(3) C/D-C(2) B/C-B-A(1)
12/05/2011	25.1	±13.4	34.9	±23.4	D(4) C-B-A(2)
20/05/2011	49.4	±16.1	54.5	±16.9	B(4) B/C-A(2) D-C(1)
26/05/2011	31.1	±12.2	28.4	±9.0	B(5) A(2) I-C-B/C(1)
01/06/2011	40.4	±11.2	61.2	±11.4	D(5) I-C(2) A(1)
10/06/2011	48.7	±17.9	65.8	±17.1	D-A(3) C/D-B/C-B(1)
17/06/2011	34.6	±11.3	45.8	±16.4	C-B-A(3) B/C(1)
22/06/2011	52.4	±20.1	72.4	±18.7	C(4) D-C/D(2) B-A(1)
29/06/2011	56.6	±14.3	83.3	±23.7	B(3) C-C/D(2) D-A-A/B(1)
07/07/2011	45.0	±10.9	57.0	±15.8	C-A(3) B(2) D-C/D(1)
14/07/2011	56.3	±16.3	59.3	±24.6	C(4) D-A(2) I-B/C(1)
22/07/2011	40.7	±9.6	56.3	±22.1	A(4) C(3) D(2) B/C(1)
02/08/2011	61.6	±20.7	81.9	±21.6	C(3) D-B-A(2)D/I(1)
10/08/2011	63.5	±16.9	78.6	±34.0	I-D-B(3) C/D(1)
19/08/2011	86.1	±28.5	83.4	±16.6	C(4) D(3) S(2) I(1)
29/08/2011	54.5	±17.9	69.8	±17.4	C(3) S-I-D(2) B(1)
08/09/2011	55.8	±16.6	76.3	±22.7	C(4) D(3) B(2) I(1)
15/09/2011	48.1	±16.1	72.9	±46.7	C(4) D(3) B(2) I(1)
22/09/2011	51.4	±20.0	73.9	±18.1	C(4) D(3) I(2) B/C(1)
30/09/2011	47.9	±17.4	76.0	±30.9	S-I-D-B(2) C-D/I(1)
06/10/2011	41.5	±21.2	55.1	±30.5	C-B-A(3) D(1)

The first cells in WT were observed again on 20 May, but only in few samples. WT cells was started in both CW and OW on 1 June, with CW tracheids already in lignification (Fig. 4.5a). Cell differentiation

completed on 22 September (DOY 265) in OW (Fig. 4.5d) and two weeks later on 6 October (DOY 279) in CW (Fig. 4.4). Results on the length of the growing season (2011) indicate that the active growth period was longer in CW (154 days) than in OW (140 days).

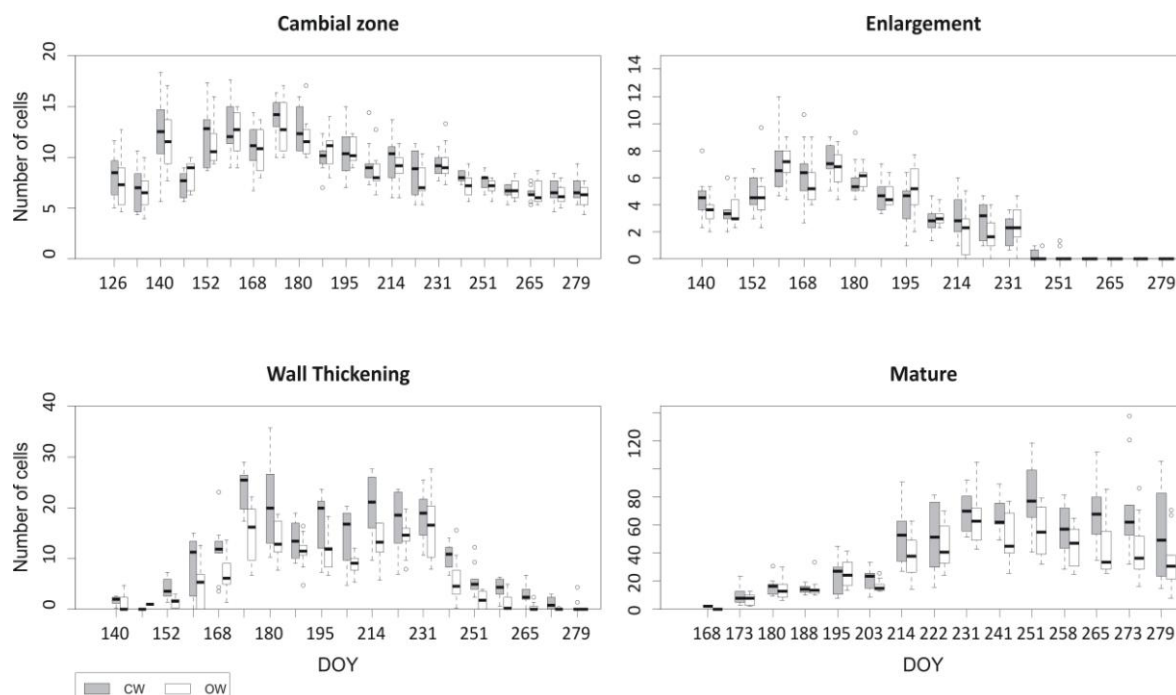
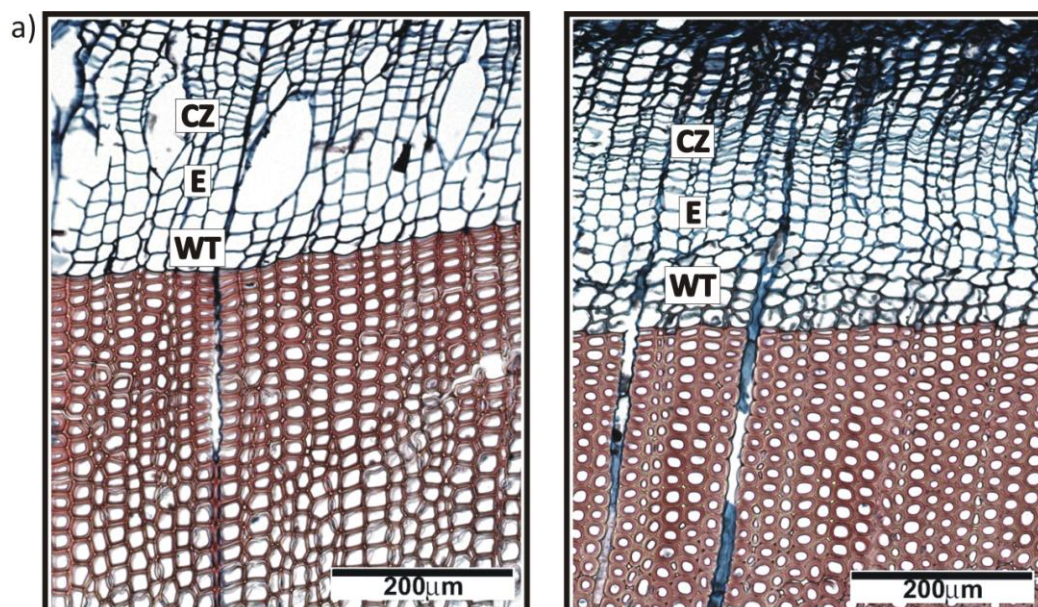
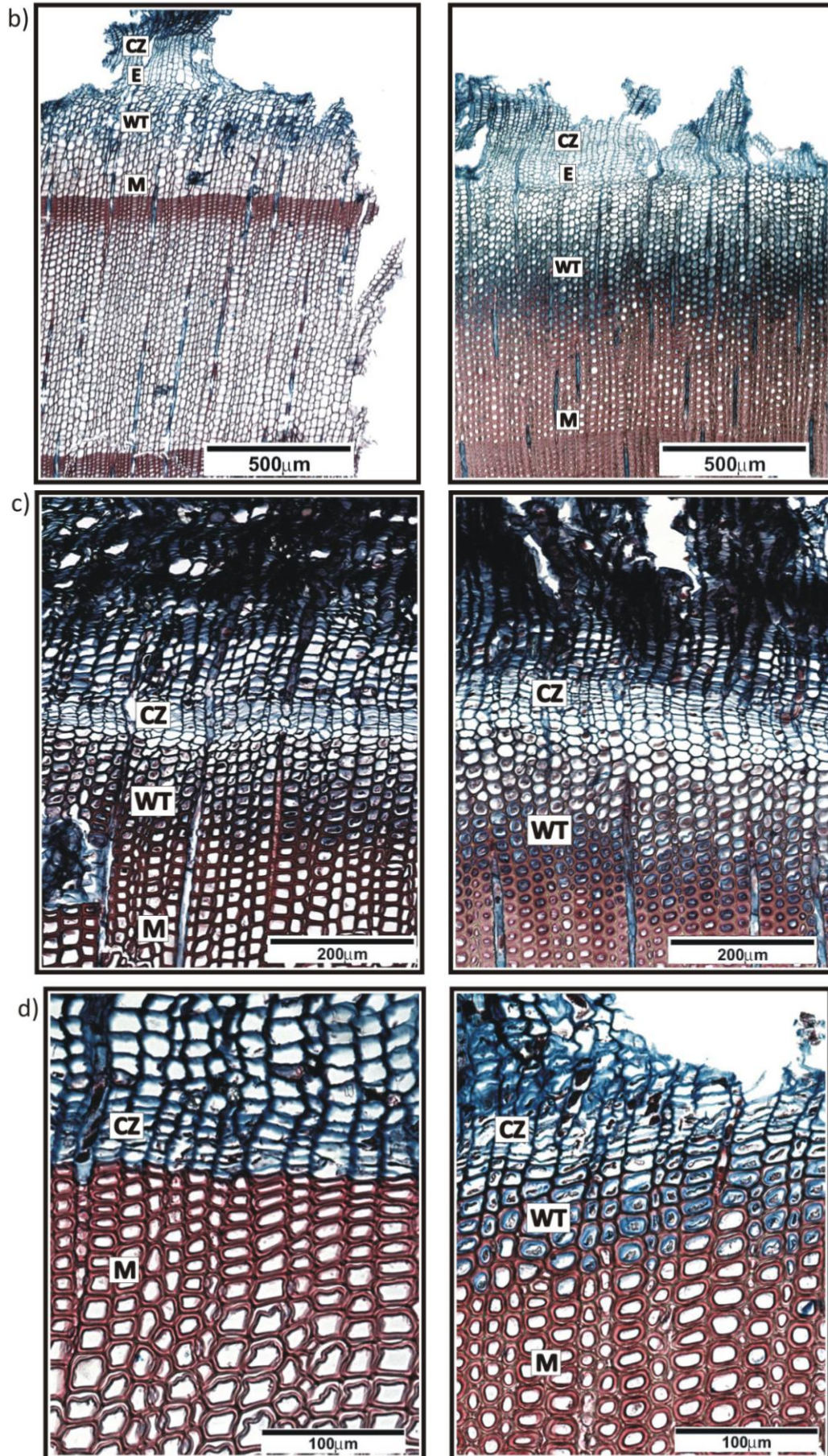


Fig. 4.4 Boxplot of the number of cells (mean of three radial files and of 10 individuals) in CW and OW during the four different phenological phases.

Fig. 4.5 Visual comparison between OW (left) and CW (right) formation. Images were taken in the same stem during particular dates: a) 1 June, start of cell wall thickening in CW and OW and lignification only in CW; b) 22 June, (maximum rate of cells production) different progress of CW and OW formation; c) 10 August, longer and higher lignification of the outer region of the secondary cell wall (S_2L) in CW; d) 22 September, end of xylogenesis only in OW





4.3.3 Xylem differentiation

While for CZ and E phase the number of cells were slightly different between CW and OW, for the WT phase this difference was higher, with 2.5 times more cells in CW than in OW. The secondary cell wall was thicker in CW than in OW, and the wall thickness was used for classifying CW severity (Table 4.1). In the 2010 growth ring of 79 samples out of 210 (38%) were characterized by relative (D) to very (S) thick secondary wall, 80% of which presented a number of cells higher than the mean value (64). During the 2011 growing season, M phase began on 22 June (DOY 173) and reached the maximum values of 217 cells in CW and 104 in OW. Out of 140 samples of CW with mature tracheids, 67% presented the degree of CW severity between D and S. The formation and lignification of $S2_L$ was clearly distinguishable during the WT phase of CW tracheids (Fig. 4.5c). An incomplete lignification of the last latewood tracheids in 2009 was observed for 13% of the samples (ANNEX II, Fig. A2c). During the four principal phenological phases (Fig. 4.6), assessed by the standardized number of cells, E counted more cells in OW than in CW, while WT phase showed the opposite. Mature cells are less numerous in CW than in OW until August, then become more numerous in CW than in OW.

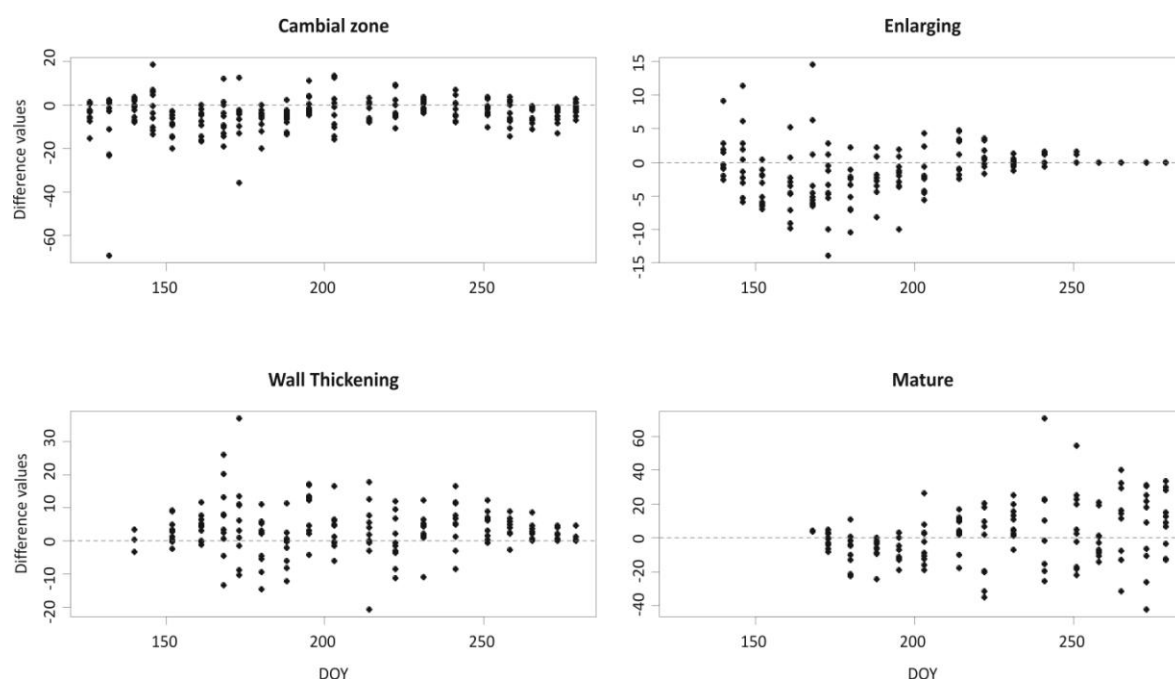


Fig. 4.6 Difference values obtained by subtracting the standardized number of cells of the OW to that of the CW during the four principal phenological phases. Note: the raw number of cells was standardized by dividing by previous ring cell count and multiplying by 100.

4.3.4 Critical dates and growing dynamic.

Results obtained from the analysis of standardized values confirmed the timings defined by using the raw number of cells. Critical data and duration of CW and OW formation (obtained averaging the standard number of cells of the 10 weekly mountain pine with the same DOY) showed higher differences in WT and M phases than E phase (ANNEX II, Table A2). These results were confirmed from the statistical test conducted on the duration of CW and OW formation, where it was hypothesized the equality between the statistical parameters of CW and OW phenological phases (ANNEX II, Fig. A2b). The WT phase began generally 2 days earlier in CW than in OW and ceased 20 days later, while the M phase began simultaneously in both wood types. Total duration of xylogenesis was also around 20 days longer in CW than in OW (ANNEX II, Fig. A2a). Despite these differences and the large variability of the number of cells among the sampled individuals, the fitting of Gompertz function yielded significant results, with r^2 of 0.96 for CW and 0.97 for OW (Fig. 4.7).

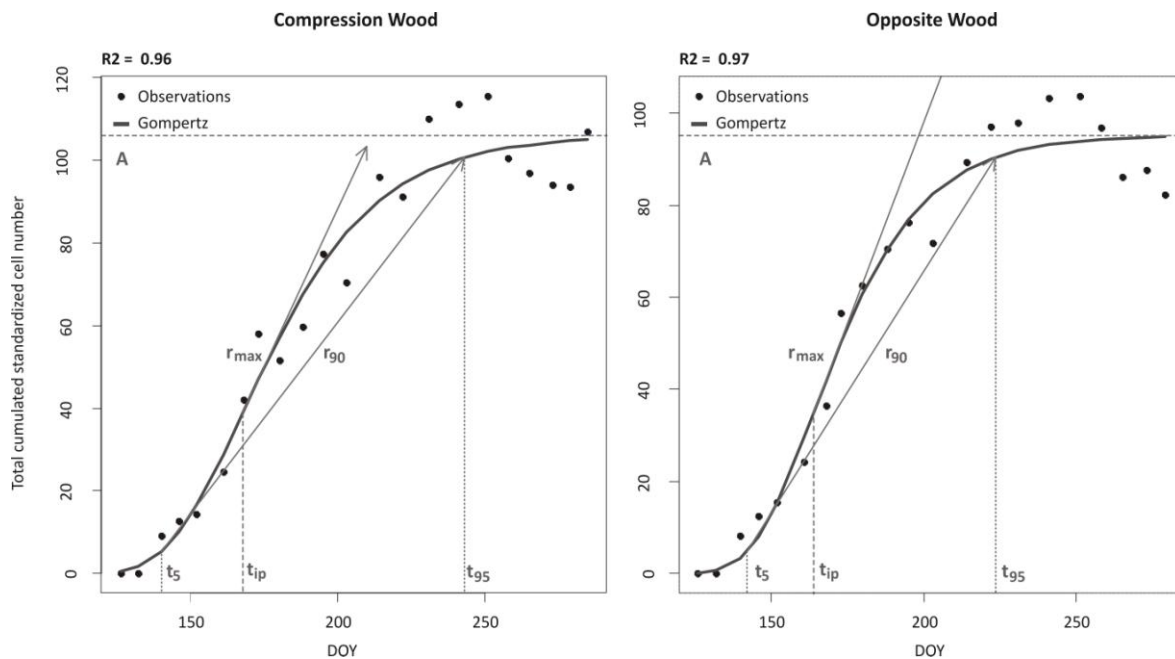


Fig. 4.7 Total number of xylem cells produced during the growing season, displayed for Compression Wood (CW) and Opposite Wood (OW). Points represent standardized data (mean of three radial files and of 10 individuals) while bold lines represent the fitted Gompertz function. Function parameters and characteristic biological parameters of wood formation are listed in Table 4.3. Note: The raw number of cells was standardized by dividing by previous ring cell count and multiplying by 100.

Mean and maximal rates of cells production were higher in CW than in OW. Date at which 5% of the cells were produced (t_5) and date at which the cell production was maximal (t_{ip}) were both slightly different (only 2 and 4 days respectively), reaching the maximal rate of tracheids production at

around the middle of June (as in the raw data) in both CW and OW. The date at which 95% of the cells were produced t_{95} was highly different between CW and OW, being delayed of 20 days in CW (31 August) in comparison with OW (11 August).

4.3.5 *Climatic influence on wood formation.*

The temperature trend showed, generally, small differences between air and stem temperatures. Two exceptional periods, however, should to be carefully considered: the last ten days of May and September. The first one was characterized by a sharp and sudden decrease in temperature, 9.5 °C (between 15 and 16 May), contemporaneously with snowy precipitation. The snow cover affected the stem temperature during the next eight days (Fig. 4.2). The second period was characterized by a sharp decrease of temperature, 10 °C (between 19 and 20 September), which affected faintly stem temperature until the end of records.

4.4 Discussions and conclusions

In mountain pine the growth rings in the CW were generally wider than those in the OW, because of the higher number of cells detected and their thicker cell wall. Hartig (1901) pointed out that the formation of CW and eccentric growth occurs simultaneously in the vast majority of cases. Other authors (Westing 1961; Riech and Ching 1970) however, stated that radial growth promotion is influenced by compressive stress on the lower side of a leaning stem, while CW formation is correlated with the gravitational stimulus. Duncker and Spiecker (2008) studied the relation of compression wood formation and eccentric radial growth and concluded that a common initiating environmental factor can be assumed for both phenomena. Results of the present study showed that eccentric growth in mountain pine is strictly connected to the higher amount of CW cells and that the radial growth began simultaneously in CW and OW, in contrast with the one-side radial growth promotion (Westing 1961; Riech and Ching 1970).

Data standardization put in evidence the relatively lower proportion of CW cells in the E phase in comparison with OW (Fig. 4.6). An opposite relation was observed considering the cells in the WT and M phases. In absolute terms, however, it should be reminded that there was a larger amount of cells in CW (on average 2.5 times that in OW), and that the mean rates of tracheids production was higher in CW (Table 4.3). This indicates that the time spent for each cell to pass through the E phase was, therefore, less in CW than in OW. The opposite was true as regard to the number of cells in the WT phase. In this case, we also observed that the proportion of mature cells gets relatively larger for

CW only after the middle of August (Fig.4.6), reaching 95% of cells production at the end of August (Table 4.3).

Table 4.3 Parameters of the Gompertz functions (a, b, k), biological parameters of the growing seasons (t_5 , t_{ip} , t_{95} , Dt_{90} , r_{max} , r_{90}) and the corresponding modelling efficiency (r^2) obtained through the analysis of standardized values of CW and OW. Gompertz function is illustrated in Fig. 4.7.

	a	b	k	t_5	t_{ip}	t_{95}	Dt_{90}	r_{max}	r_{90}	r^2
CW	106.0042	6.614264	0.039424	139.9423	167.7728	243.1127	103.1704	1.537406	0.92472	0.962674
OW	95.12285	8.21522	0.050057	142.1983	164.117	223.4532	81.25491	1.751685	1.053605	0.969546

Note: Raw number of cells were standardized by dividing by previous ring cell count and multiplying by 100.

t_5 : date at which 5% of the cells are produced;

t_{ip} : date at which the cell production is maximal;

t_{95} : date at which 95% of the cells are produced;

Dt_{90} : time elapsed between t_5 and t_{95} ;

r_{max} : maximal rate of cell production;

r_{90} : mean rate of cell production computed between t_5 and t_{95} .

(Rathgeber 2012)

Considering that the WT phase was 20 days longer for CW (ANNEX II, Fig. A2a) and that the CW cells remained longer in the WT phase (Fig. 4.6), it may be hypothesized that CW cells need definitively more time to build the cell wall than cells in OW. The longer time spent for a more numerous number of CW cells in the WT phase might represent a strategy for those cells to mature, while having thicker cell walls. To best reach this goal, the CW expanded the growing season of two additional weeks, until 6 October (DOY 279), with the risk that, if the weather condition deteriorated, the maturation could not be fully completed, as it occurred in 2009 for 13% of the sampled individuals (ANNEX II, Fig. A2c). Many authors studied the differentiation phase in late autumn, demonstrating the incomplete lignification of secondary walls of the last-formed tracheids for several conifers (Murmanis and Sachs 1969; Nix and Villiers 1985; Donaldson 1991, 1992; Gričar et al. 2005). Donaldson (2001) suggested that lignification is sensitive to environmental conditions, especially temperature, which seems to play an important role in slowing down this process. Gričar et al. (2005) stated that duration of cambial activity is longer in trees with broad annual rings than in trees with narrow rings. They demonstrated that, in trees with broader annual rings, the final steps of differentiation of last-formed tracheids still continues during autumn, and these were finished prior to winter. This result agrees with our findings, although Gričar et al. (2005) made no reference to the presence of compression wood, although the presence of compression wood was clearly evident in the published pictures.

Lupi et al. (2010) demonstrated that the onset of xylogenesis would affect the number of cells produced by the cambium, which, in turn, would influence the ending of cell differentiation. In particular, a larger amount of cells produced by the CZ would increase the time spent in the differentiation and maturation phases, delaying the ending of wood formation (Lupi et al., 2010; Rossi et al., 2011). Rossi et al. (2012) found that higher accumulations of undifferentiated cells in cambium need more time to undergo differentiation, delaying the ending of cell enlargement and wall thickening and lignification in late summer and extending the length of xylogenesis. Also, even with greater cell production, xylem differentiation would not be able to occur faster. Our results agree with these hypotheses, adding new insights about the timing in cell differentiation. The onset of xylogenesis was considered directly related with the number of cells produced, but in mountain pine though the onset was the same, the amount of produced cells between CW and OW was totally different. Certainly the wall thickening requires longer period to complete cell maturation, but if in the same stem a different number of cells were produced with slightly different time (only two weeks), tracheids lignification would probably occur in different ways. In this sense, potassium permanganate staining has been widely used in electron microscopy as an indicator for lignin distribution in different morphological regions of woody cell walls (e.g. Maurer and Fengel 1991; Donaldson 1992; Singh and Donaldson 1999; Schmitt and Melcher 2004). Gričar et al. (2005), which could be of help in identifying the stages of differentiation in the last-formed tracheids.

In order to exactly quantify timings of CW formation it would be necessary to follow cell formation and differentiation along the same stem of mountain pine. In this case the number of cells should increase progressively both in CW and OW and comparing the progress of the development of the two wood types would be possible. However, the structure of mountain pine would bias sampling many (42 in our case) microcores from the same stem, without causing consecutive wound reactions. Moser et al. (2010) demonstrated that the length of growing season is directly correlated with the trend of air temperature. Rossi et al. (2010) and Moser et al. (2010) observed a reduction in the period and amount of xylem production along elevation gradients. In our case, at 1900 m a.s.l., mountain pine growing season ended during the first half of October; similar results were found by Rossi et al. (2008a) for *Larix decidua* Mill. and *Picea abies* (L.) at 2080 m a.s.l. in the eastern Italian Alps. Mountain pine is a very sensitive species to changing environmental conditions, growing above the timberline. Dendrochronological studies conducted on climate-growth relationship of mountain pine demonstrated that its growth is mostly influenced by spring maximum temperature (Pelfini et al. 2006, Palombo et al. submitted), and by October maximum temperature and summer precipitation on the Majella massif (Palombo et al. submitted). In agreement with dendrochronological investigations, and following the predicted climatic trend of increasing temperature (Brunetti et al. 2006) it may be supposed an elongation of mountain pine growing

season in the studied environment, these months representing the beginning and the end of the growing season. Rossi et al. (2012) found that the successive phenological phases of cell production and differentiation are closely interconnected by complex relationships of cause and effect, and suggested that xylogenesis could have a precise and recognizable framework. Our findings strengthen this theory because different phenological phases with different timings occurred along the same stem of mountain pine trees, as if these phases were driven by two different stimuli.

Acknowledgements We thank the Majella National Park for access to sampling sites and permission to core trees. We are grateful to Luciano Schiazza, Giuseppe D’Ascanio, Gianni Blasioli, Roberto Iezzi and the workers from the Corpo Forestale dello Stato (Comando stazione di Caramanico Terme) for their support in fieldwork, and Holger Gärtner and Fritz Schweingruber for valuable suggestion and fruitful discussion.

References

- Anfodillo T., Deslauriers A., Menardi R., Tedoldi L., Petit G., Rossi S. 2012. Widening of xylem conduits in a conifer tree depends on the longer time of cell expansion downwards along the stem. *Journal of experimental botany* 63: 837–45.
- Brunetti M., Maugeri M., Monti F., Nanni T. 2006. Temperature and precipitation variability in Italy in the last two centuries from homogenised instrumental time series. *Int J Climatol* 26: 345–381.
- Côté W.A., Day A.C., Timell T.E. 1968. Studies on compression wood VII. Distribution of lignin in normal and compression wood of tamarack *Larix laricina* (Du Roi, K.Koch). *Wood Sci. Technol.* 2: 13–37.
- Donaldson L.A. 1991. Seasonal changes in lignin distribution during tracheid development in *Pinus radiata*. *Wood Science and Technology* 25: 15–24.
- Donaldson L.A. 1992. Lignin distribution during latewood formation in *Pinus radiata*. *IAWA Bulletin* 12: 381–387.
- Donaldson L.A. 2001. Lignification and lignin topochemistry - an ultrastructural view. *Phytochemistry* 57: 859–873.
- Donaldson L.A., Singh A.P., Yoshinaga A., Takabe K. 1999. Lignin distribution in mild compression wood of *Pinus radiata* D. Don. *Can. J. Bot.* 77: 41–50.
- Donaldson L.A. and Turner J.C.P. 2001. The influence of compression wood and microfibril angle on the occurrence of distortion in window frames made from radiata pine (*Pinus radiata*). *Holz Roh Werkst.* 59: 163–168.
- Donaldson L.A., Grace J.C., Downes G. 2004. Within tree variation in anatomical properties of compression wood in radiata pine. *IAWA J.* 25: 253– 271.

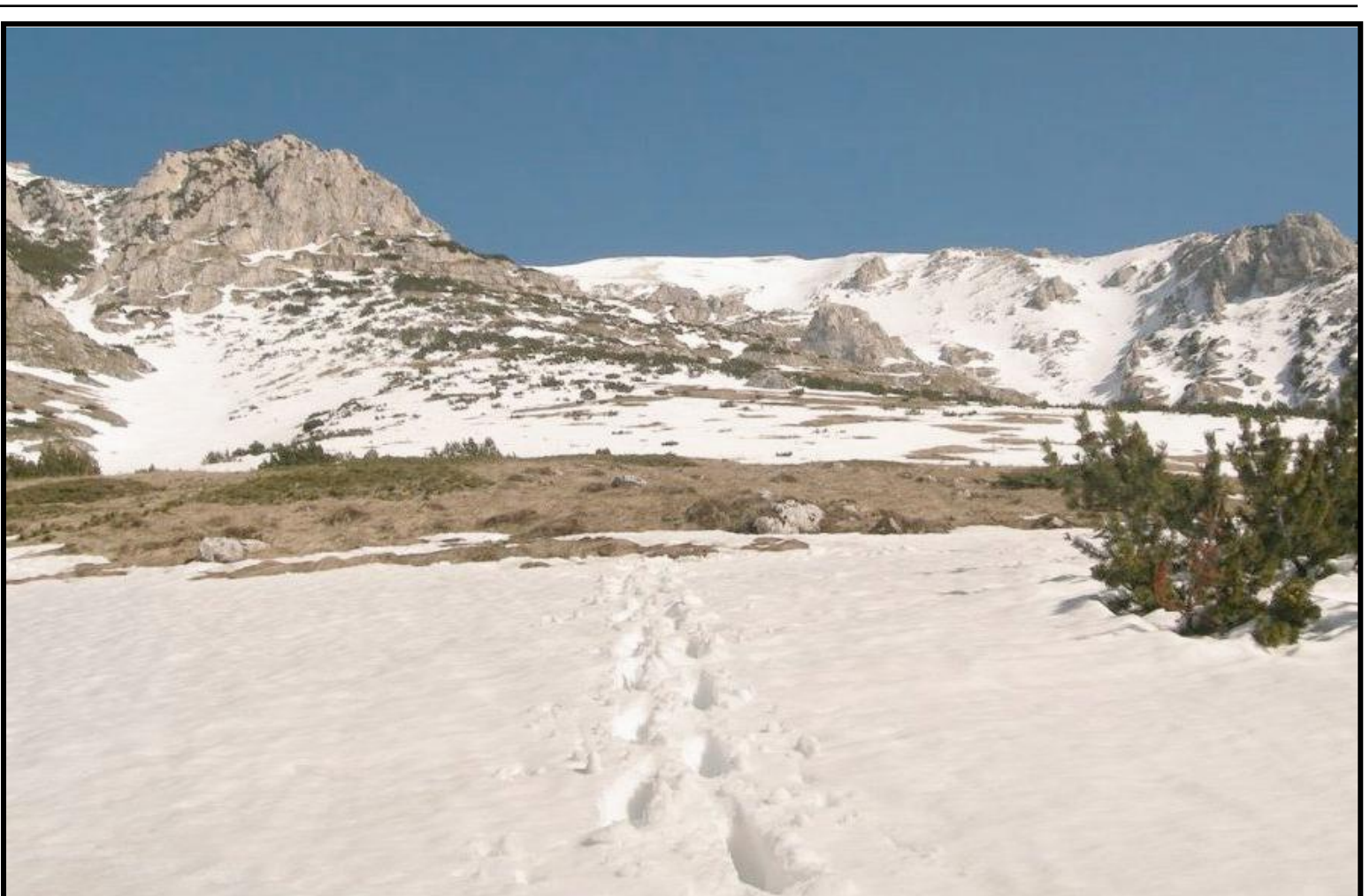
- Duncker P. and Spiecker H. 2008. Cross-sectional compression wood distribution and its relation to eccentric radial growth in *Picea abies* [L.] Karst. *Dendrochronologia* 26: 195–202.
- Eilmann B., Zweifel R., Buchmann N., Fonti P., Rigling A. 2009. Drought-induced adaptation of the xylem in Scots pine and pubescent oak. *Tree physiology* 29: 1011–20.
- Fonti P., Von Arx G., García-González I., Eilmann B., Sass-Klaassen U., Gärtner H., Eckstein D. 2010. Studying global change through investigation of the plastic responses of xylem anatomy in tree rings. *The New phytologist* 185: 42–53.
- Fonti P., Heller O., Cherubini P., Rigling A., Arend M. 2013. Wood anatomical responses of oak saplings exposed to air warming and soil drought. *Plant Biology* 15 (Supl. 1): 210–219.
- Frankenstein C, Eckstein D, Schmitt U. 2005. The onset of cambium activity – a matter of agreement? *Dendrochronologia* 23: 57–62.
- Galle A., Esper J., Feller U., Ribas-Carbo M., Fonti P. 2010. Responses of wood anatomy and carbon isotope composition of *Quercus pubescens* saplings subjected to two consecutive years of summer drought. *Annals of Forest Science* 67: 809–809.
- Gardiner B. and Macdonald E. 2005 Compression wood in conifers – the characterization of its formation and its relevance to timber quality, in European Union – *Framework Programme FP5 – Quality of Life and Management of Living Resources*, QLRT-2000-00177, pp. 376
- Gričar J., Čufar K., Oven P., Schmitt U. 2005. Differentiation of terminal latewood tracheids in silver fir trees during autumn. *Annals of botany* 95: 959–65.
- Gričar J., Zupančič M., Čufar K., Koch G., Schmitt U., Oven P. 2006. Effect of local heating and cooling on cambial activity and cell differentiation in the stem of Norway spruce (*Picea abies*). *Annals of botany* 97: 943–951.
- Gričar J., Zupančič M., Čufar K., Oven P. 2007. Regular cambial activity and xylem and phloem formation in locally heated and cooled stem portions of Norway spruce. *Wood Science and Technology* 41, 463–475.
- Hartig R. 1901. Holzuntersuchungen. Altes und Neues. Springer, Berlin, p. 99.
- Lehner G. and Lütz C. 2003. Photosynthetic functions of cembran pines and dwarf pines during winter at timberline as regulated by different temperatures, snowcover and light. *Journal of plant physiology* 160: 153–66.
- Lupi C., Morin H., Deslauriers A., Rossi S. 2010. Xylem phenology and wood production: resolving the chicken-or-egg dilemma. *Plant, Cell and Environment* 33: 1721–1730.
- Maurer A. and Fengel D. 1991. Elektronenmikroskopische Darstellung von strukturellen Einzelheiten in Nadelholz-Zellwänden anhand sehr dünner Ultramikrotomschnitte. *Holz Roh-Werkstoff* 49: 53–56.

- Moser L., Fonti P., Buentgen U., Franzen J., Esper J., Luterbacher J., Frank D. 2010. Timing and duration of European larch growing season along altitudinal gradients in the Swiss Alps. *Tree Physiology* 30, 225–233.
- Murmanis L. and Sachs I. 1969. Seasonal development of secondary xylem in *Pinus strobus* L. *Wood Science and Technology* 3: 177–193.
- Nanayakkara B., Manley-Harris M., Suckling I.D., Donaldson L.A. 2005. Chemical characterisation of compression wood in *Pinus radiata*. In: *13th International Symposium on Wood and Pulping Chemistry*, Auckland, New Zealand.
- Nanayakkara B., Manley-Harris M., Suckling I.D., Donaldson L.A. 2009. A quantitative chemical indicator of compression wood severity. *Holzforschung* 63: 431–439.
- Nix L.E. and Villiers K. 1985. Tracheid differentiation in southern pines during the dormant season. *Wood and Fibre Science* 17: 397–403.
- Palombo C., Battipaglia G., Cherubini P., Chirici G., Garfi V., Lasserre B., Lombardi F., Marchetti M., Tognetti R. *Submitted*. Warming-related growth responses at the southern limit distribution of mountain pine (*Pinus mugo* Turra ssp. *mugo*).
- Pelfini M., Leonelli G., Santilli M. 2006. Climatic and Environmental Influences on Mountain Pine (*Pinus Montana* Miller) Growth in the Central Italian Alps. *Arctic, Antarctic, and Alpine Research* 38, 614–623.
- Prislan P., Schmitt U., Koch G. 2011. Seasonal ultrastructural changes in the cambial zone of beech (*Fagus sylvatica*) grown at two different altitudes. *IAWA Journal* 32: 443–459.
- R Development Core Team. 2007. R: A Language and Environment for Statistical Computing. R Development Core Team, Vienna, Austria.
- Rathgeber C.B.K. 2012. Cambial activity and wood formation: data manipulation, visualisation and analysis using R. *R package version 1.4-1*.
- Rathgeber C.B.K., Longuetaud F., Mothe F., Cuny H., Le Moguedec G. 2011. Phenology of wood formation: data processing, analysis and visualisation using R. *Dendrochronologia* 29:139–149.
- Riech F.P. and Ching K.K. 1970. Influence of bending stress on wood formation of young Douglas-Fir. *Holzforschung* 24: 68–70.
- Rossi S., Deslauriers A., Morin H. 2003. Application of the Gompertz equation for the study of xylem cell development. *Dendrochronologia* 21: 33–39.
- Rossi S., Anfodillo T., Menardi R. 2006a. Trephor: a new tool for sampling microcores from tree stems. *IAWA Journal* 27: 89–97.
- Rossi S., Deslauriers A., Anfodillo T. 2006b. Assessment of cambial activity and xylogenesis by microsampling tree species: an example at the Alpine timberline. *IAWA Journal* 27: 383–394.

- Rossi S., Deslauriers A., Gričar J., Seo J.W., Rathgeber C.B.K., Anfodillo T., Morin H., Levanic T., Oven P., Jalkanen R. 2008a. Critical temperatures for xylogenesis in conifers of cold climates. *Global Ecology and Biogeography* 17: 696–707.
- Rossi S., Deslauriers A., Anfodillo T., Carrer M. 2008b. Age-dependent xylogenesis in timberline conifers. *The New phytologist* 177: 199–208.
- Rossi S., Morin H., Tremblay M.J. 2010. Growth and productivity of black spruce (*Picea mariana*) belonging to the first cohort in stands within and north of the commercial forest in Quebec, Canada. *Annals of Forest Science* 67: 807.
- Rossi S., Morin H., Deslauriers A., Plourde P.Y. 2011. Predicting xylem phenology in black spruce under climate warming. *Global Change Biology* 17: 614–625.
- Rossi S., Morin H., Deslauriers A. 2012. Causes and correlations in cambium phenology: towards an integrated framework of xylogenesis. *Journal of experimental botany* 63: 2117–26.
- Schmitt U. and Melcher E. 2004. Section staining with potassium permanganate for transmission electron microscopy: a useful tool for lignin localisation. In *Wood fibre cell walls: methods to study their formation, structure and properties*, Schmitt U. et al. (eds.). Uppsala, Sweden: Swedish University of Agricultural Sciences: 105–118.
- Schweingruber F.H. 1996. Tree rings and environment - dendroecology. Bern, Switzerland: Paul Haupt, pp. 609.
- Schweingruber F.H. 2006. Wood structure and environment. Berlin Heidelberg, New York, NY, USA: Springer-Verlag.
- Singh A.P. and Donaldson, L.A. 1999. Ultrastructure of tracheid cell walls in radiata pine (*Pinus radiata* D. Don) mild compression wood. *Can. J. Bot.* 77: 32–40.
- Timell T.E. 1973. Ultrastructure of the dormant and active cambial zones and the dormant phloem associated with formation of normal and compression woods in *Picea abies* (L.) Karst. *Technical publication n.96*. State University of New York, College of Environmental Science and Forestry. Siracuse, New York, pp.93.
- Timell T.E. 1986. The structure of compression wood, Chemical properties of compression wood. In *Compression Wood in Gymnosperms* Volume 1, Springer-Verlag, Berlin, pp. 81.
- Westing A.H. 1961. Changes in radial symmetry in the leaders of eastern white pine following inclination. *Journal of Forestry* 56: 17–19.
- Yumoto M., Ishida S., Fukazawa K. 1983. Studies on the formation and structure of compression wood cells induced by artificial inclination in young trees of *Picea glauca*. IV. Gradation of the severity of compression wood tracheids. *Research Bulletins of the College Experiment Forests Hokkaido University* 40:409

FIFTH CHAPTER:

Summary and Conclusion



“Mediterranean forests are characterised by a remarkable set of features that make them naturally and aesthetically attractive, on the one hand, but also quite fragile, on the other, therefore calling for careful strategies for their conservation and management” Scarascia-Mugnozza et al. (2000). Although the main patterns of climate-growth relationships are well known, the large seasonality and year-to-year variability that characterize precipitation and temperature in the Mediterranean region and the different site-dependent seasonality of tree growth in forests from this area may make very difficult to determine the response times of tree growth to the precipitation deficit and increasing temperatures. Here the response of mountain pine to varying growth conditions through time was assessed at the stand, individual and tissue levels on the Majella massif of Central Apennines.

Mountain ecosystem, actually, are threatened by many factors, among which the principal ones are climate, land-use and their interaction. Although their impacts cannot as yet be predicted with certainty, mountain systems are particularly sensitive to changes in climate (Beniston 2000), especially in the Mediterranean Basin identified as one of the areas that is most vulnerable to the predicted climate change Giorgi (2006). Mediterranean mountains are under threat from climate change, which affects directly or indirectly different key features, such as biodiversity, snow cover, glaciers, run-off processes and water availability (Vogiatzakis 2012). As well as, during the twentieth century, one of the most geographically extensive land-cover and landscape changes globally was woody plant invasion of grass-dominated ecosystems, because of the declines of traditional practices based on small-scale (i.e. agriculture, pastoralism and forest resource utilization in marginal areas, generally situated in the mountains - Chauchard et al. 2007). The changes that are involving the high-altitude ecosystems within the Mediterranean Basin represent an important proxy for assessing the ongoing global changes and, on the contrary, forecasting future climate and land-use changes will allow predicting possible scenarios of ecosystems evolution. In the context of a National Park, this knowledge is crucial for planning conservation strategies and locally tailored sustainable management practices. Identifying the principal factors that influence the development dynamics of the dominant species at treeline on the Majella massif, Central Apennines, this study contributes to better predict the future behaviour of these ecotonal ecosystems.

The first chapter showed a considerable expansion of mountain pine krummholz during the last fifty years (from 693 ha in 1954 to 1423 ha in 2007). This increase was mainly concentrated during the period 1954-1991, due to a downward expansion (3 m per year) and a marked ability of this species to recolonize large areas previously grazed or cut (forest cover increase of 2.1%), simultaneously with the abandonment of summer pastoralism. This result confirms the importance of human activities in shaping the treeline ecotone of Central Apennines and supposes an increasing trend of krummholz expansion directly related to the establishment of the Majella National Park on 5 June 1995, and then to the greater number of constraints that favours the abandonment of traditional practices. It

was also observed an upward expansion of mountain pine krummholz of 5 m per year, which, on the contrary, occurred during the last twenty years (1991-2007). In this case, the elevation increase of krummholz vegetation involved small patches in remote and inaccessible areas (Fig. 2.3), indicating the colonization by mountain pine of areas where, probably, the climatic conditions only recently have become suitable for its growth. For this reason it was necessary to identify the climatic parameters that more strongly influence the mountain pine growth.

The third chapter demonstrated that mountain pine growth is particularly affected by climatic parameters during the most important months of its growing season. Maximum temperatures were found to positively affect tree-ring growth during spring months and October, while negatively during summer months, especially at higher altitude. On the other hand, total precipitation resulted very important both in April, when its influence on tree-ring growth, particularly at lower altitude was negatively oriented, and during summer months when it exerted a positive influence on tree-ring development.

In order to better interpret the dendroclimatological results, and then to understand how specific climatic factors influence the cellular division, growth and maturation that collectively define tree-ring characteristics, the timing and duration of the growing season was usefully assessed by monitoring the wood formation through anatomical observations.

The fourth chapter revealed that maximum temperatures represent the critical factor for the ring width formation, influencing positively tree growth in spring months, i.e. during the beginning of the growing season when the cell division and enlargement processes are in progress. In addition, the positive correlations found between summer precipitation and tree-ring formation are very important for understanding the wood development dynamics, both because the study area is characterized by Mediterranean climatic and karst substrate, and for the concurrent phase of maturation of tracheids, then requiring energy for their lignification. The maturation phase in mountain pine resulted also the most important phenological phase, this species being characterized by the high percentage of compression wood. In fact, a different timing in the formation of compression wood and opposite wood within the same stem was observed, compression wood needing a longer and greater lignification of its tracheids.

According with the projections of the future climate change, the Mediterranean Basin is expected to experience greater increases in temperature and aridity than are expected in other regions (IPCC, 2007). Already observed changes in climatic trends include a decrease in precipitation in the Mediterranean area for the period 1961–1990 (De Luís et al. 2001; Peñuelas et al. 2002), with increasing intensity of annual rainfall patterns (De Luís et al. 2001). Precipitation is further expected to decrease in summer and autumn, but to increase in winter, resulting in higher intensity of single precipitation events, reduction of soil moisture (Déqué et al. 1998) and an overall decrease in water

availability due to increased evapotranspiration. This climatic scenario, nevertheless, would be favourable for an elongation of mountain pine growing season, which would be able to start growing earlier in the season, from one side, though this could compromise wood formation during drier summer periods, on the other. Higher temperatures could not only affect the length of the growing season, but might also allow mountain pine growth in areas previously unfavourable. These last aspects may have important influences on the distribution of rare and endemic species within the alpine belt of the Majella massif.

In this context, the Majella National Park was included in the 'Global Observation Research Initiative in Alpine Environments' program (project UE-GLORIA 2001–2003 – Co-ordinator Prof. Grabherr, University of Vienna). Stanisci et al. (2005), on the basis of the results obtained in this project, hypothesized that climatic warming projections for the year 2100 (+3.5 °C) will remarkably affect the alpine ecosystems of the central Apennines, according to what already observed for similar environments on the Alps (Wanner and Beniston 1995; Theurillat et al. 1998; Körner 1999; Pauli et al. 2001; Theurillat and Guisan 2001). The first process hypothesized would concern the rise from below of species belonging to bioclimatic belts at lower altitudes, followed by an increase of species richness in habitats of the alpine belt (Guisan et al. 1995; Gottfried et al. 1999; Grabherr et al. 2001). Results obtained by Stanisci et al. (2005) suggest, for the alpine belt of the Majella massif, the initial arrival of thermophilous species from below on the eastward exposures, whereas the northward exposures, with a shorter frost-free period, would be the most conservative, showing greater inertia toward the invasive process. Recently, a more comprehensive study was conducted by Gottfried et al. (2012) based on the standardized and multiple-scale dataset of GLORIA project for European mountain systems (Pauli et al. 2004) for the period 2001–2008. In this study, which covered the treeline ecosystems from 60 summit sites in all major European mountains, including the Majella massif, the ongoing climate change was shown to gradually transform mountain plant communities. These authors provided evidence that the more cold-adapted species would decline and the more warm-adapted species increase (thermophilization). They also indicated that this process in mountain plant communities mirrors the degree of recent warming, being more pronounced in areas where the increase in air temperature has been higher. Again, the observed transformation suggests a progressive decline of cold mountain habitats and their biota.

Because of all these threats, the Majella National Park authorities ask for guidelines for an active forest management that curbs global warming and land-use change effects. Sustainable forest management practices are desirable to monitor and control the excessive expansion of mountain pine, without compromising its natural diffusion in the Majella massif, in species-rich grasslands at high elevation. However, mountain pine population of the Majella massif is an isolated patch within

the Mediterranean basin. The dispersal ability of this relictual vegetation is reduced compared to its distance from potentially suitable sites for its growth. For this reason it is important to avoid the action of invasive species, which could threaten the presence of mountain pine krummholz in the Majella massif. Among the possible actions towards the sustainable management of mountain pine krummholz some principles for forest species adaptation are suggested: (1) preservation of the pool of species and genetic variability of this population, increasing the cultivation *ex situ*, in the alpine botanical gardens already in place, and improving the seed bank. (2) Networking of protected areas or natural ecosystems by creating ecological corridors or by maintaining appropriate ecological components in associated areas to allow natural migration of ecosystem elements; thanks to the three national parks located in the Abruzzo Region, very close to each other, establishing buffer zones in order to allow for ecosystem replacement would be practical. (3) Reduction of forest fragmentation caused by anthropogenic activities and restoration of degraded lands. Maintaining the current open landscape would require mowing a larger area, reconsidering the absolute restriction for grazing activities, focused more intensively on un-mown grasslands and applied early in the season, could also be useful to restore species-rich grasslands. (4) description of best management practices that will maintain or enhance adaptive capacity or resilience of high elevation ecosystems.

Future researches are necessary for assessing the role of beech forest in the treeline ecotone dynamics on the Majella massif, as well as monitoring the climatic trends. During the growing season of 2011, microcores were taken from 10 individuals of *Fagus sylvatica* (L.) and a data logger was installed to record the temperature trend at the upper limit of timberline. Establishing permanent plots within an altitudinal transect in the Majella massif would allow a comprehensive understanding of functional dynamics between adjacent ecosystems and a quantitative and qualitative analysis of the intrinsic and extrinsic factors influencing biodiversity and spatial distribution changes.

The effect of drought stress on mountain pine and other species growing at the alpine timberline, either due to freeze-thaw events or low seasonal precipitation. Frost drought during winter could cause high embolism rates unless xylem vulnerability or transpiration is sufficiently reduced to avoid critical water potential. Detailed anatomical analysis could provide new insights on the structural and functional responses of treeline vegetation to extreme climatic events, as early or late frosts and recurrent summer drought. Indeed, shifts in rainfall patterns and increasing temperatures associated with climate change are likely to cause widespread forest decline in regions where droughts are predicted to increase in duration and severity, such as the Mediterranean Basin. Possible strategic responses of these forest ecosystems to climatic change fall into four broad classes: restraint (do nothing), resilience (absorb stress), resistance (near-term ways of buying time), and realignment (long-term adaptation). Planning responses will be made challenging by the unprecedented and

unpredictable nature of future changes; robust planning approaches, like scenario planning, are necessary for all the Circum-Mediterranean forests.

References

- Beniston M. 2000. *Environmental Change in Mountains and Uplands*. Arnold.
- Chauchard S., Carcaillet C., Guibal F. 2007. Patterns of Land-use Abandonment Control Tree-recruitment and Forest Dynamics in Mediterranean Mountains. *Ecosystems* 10: 936–948.
- De Luís M., García-Cano M.F., Cortina J., Raventós J., González-Hidalgo J.C., Sánchez J.R. 2001. Climatic trends, disturbances and short-term vegetation dynamics in a Mediterranean shrubland. *Forest Ecology and Management* 147: 25–37.
- Déqué M., Marquet P., Jones R.G. 1998. Simulation of climate change over Europe using a global variable resolution general circulation model. *Climate Dynamics* 14: 173–189.
- Giorgi F. 2006. Climate change hot-spots. *Geophysical Research Letters* 33: L08707.
- Gottfried M., Pauli H., Reiter K., Grabherr G. 1999. A fine-scaled predictive model for changes in species distribution patterns of high mountain plants induced by climate warming. *Divers. Dist.* 5: 241–251.
- Gottfried M., Pauli H., Futschik A., Akhalkatsi M., Barančok P., Benito Alonso J.L., Coldea G., Dick J., Erschbamer B., Fernández Calzado M.R., Kazakis G., Krajčí J., Larsson P., Mallaun M., Michelsen O., Moiseev D., Moiseev P., Molau U., Merzouki A., Nagy L., Nakhutsrishvili G., Pedersen B., Pelino G., Puskas M., Rossi G., Stanisci A., Theurillat J.P., Tomaselli M., Villar L., Vittoz P., Vogiatzakis I., Grabherr G. 2012. Continent-wide response of mountain vegetation to climate change. *Nature Climate Change* 2: 111–115.
- Grabherr G., Gottfried M., Gruber A., Pauli H. 2001. Aspects of global change in the Alps and in the high arctic region. Long-term monitoring of mountain peaks in the Alps. In: Burga C.A. and Kratochwil A. (eds) *Biomonitoring: General and Applied Aspects on Regional and Global Scales*. Kluwer Academic Publishers, Dordrecht, The Netherlands, pp. 153–177.
- Guisan A., Holten J.I., Spichiger R., Tessier L. (eds) 1995. Potential ecological impacts of climate change in the Alps and Fennoscandian mountains. Publication hors-séries des Conservatoire et Jardin botaniques de la Ville de Genève, Département des affaires culturelles, 8. Genève, Switzerland.
- IPCC (2007). *Climate change 2007: impacts, adaptation and vulnerability. Southern Europe*. Working Group II Contribution to the Intergovernmental Panel on Climate Change, United Nations Environmental Program, Cambridge University Press, Cambridge.
- Körner C. 1999. *Alpine Plant Life*. Springer Verlag, Berlin, Germany.

- Pauli H., Gottfried G., Grabherr G. 2001. High summits of the Alps in a changing climate. In *Fingerprints of Climate Change, Adapted Behaviour and Shifting Species Range*, Walter A., Burga A. and Edwards P.J. (eds.). Kluwer Academic Publishers, Dordrecht, the Netherlands, pp. 139–149.
- Pauli, H. et al. 2004. The GLORIA Field Manual - Multi-Summit Approach (European Commission DG Research, EUR 21213, Office for Official Publications of the European Communities, European Commission).
- Peñuelas J., Filella I., Comas P. 2002. Changed plant and animal life cycles from 1952–2000 in the Mediterranean region. *Global Change Biology* 8: 531–544.
- Scarascia-Mugnozza G., Oswald H., Piussi P., Radoglou K. 2000. Forests of the Mediterranean region: gaps in knowledge and research needs. *Forest Ecology and Management* 132: 97–109.
- Stanisci A., Pelino G., Blasi C. 2005. Vascular plant diversity and climate change in the alpine belt of the central Apennines (Italy). *Biodiversity and Conservation* 14: 1301–1318.
- Theurillat J.P. and Guisan A. 2001. Impact of climate change on vegetation in the European Alps: a review. *Climatic Change* 50: 77–109.
- Theurillat J.P., Felber P., Geissler P., Gobat J.M., Fierz M., Fischlin A., Kupfer P., Schlusser A., Velluti C., Zhao G.F. 1998. Sensitivity of plant and soil ecosystems of the Alps to climate change. In *Views from the Alps. Regional Perspectives on Climate Change*, Cebon P., Dahinden U., Imboden D. and Jager C. (eds.). MIT Press, Boston, Massachusetts, pp. 225–308.
- Vogiatzakis. I.N. 2012. Mediterranean Mountain Environments. First Edition, John Wiley & Sons, Ltd., pp. 216.
- Wanner H. and Beniston M. 1995. Approaches to the establishment of future climate scenarios for the alpine region. In *Potential Ecological Impacts of Climate Change in the Alps and Fennoscandian Mountains*, Guisan A., Holten J., Spichiger R. and Tessier L. (eds.). Publication hors-série n. 8 des Conservatoire et Jardin botaniques de la Ville de Genève. Département des affaires culturelles, Genève, Switzerland, pp.87–96.

ANNEX I

Fig. A1a. Climatic diagram obtained from the average of climatic data recorded from 3 meteorological station (Pescocostanzo, 1461 m a.s.l. – Lat. 41° 53' 09"N Long. 14° 04' 14"E; S.Eufemia a Maiella, 888 m a.s.l. – Lat. 42° 07' 00"N Long. 14° 01' 08"E; Palena, 767 m a.s.l. – Lat. 41° 59' 01"N Long. 14° 08' 15"E) site in the MNP boundaries and considered the most representative of the study area. Time span: 1932 - 2010 for Mean Temperatures and 1920 - 2010 for Total Precipitation. The mean annual temperature for the period 1932-2010 is 10.2 °C and the mean total annual precipitation for the period 1920-2010 is 1140.2 mm of rain.

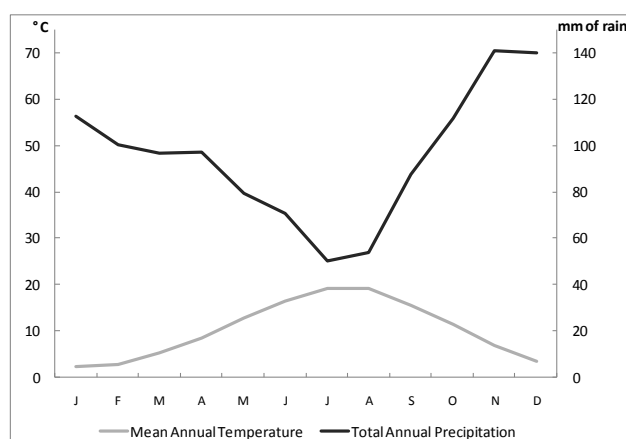


Fig. A1b. Variation and trend of maximum and minimum mean temperature from 1932 to 2010 and total precipitation from 1920 to 2010 with annual and seasonal resolution (Spring: March-May; Summer: June-August; Autumn: September-November; Winter: December_(t-1)- February). Monthly climatic data are obtained averaging records of the 3 considered meteorological station. Y-axis present the same scale of values but with different ranges on the basis of the variables and season.

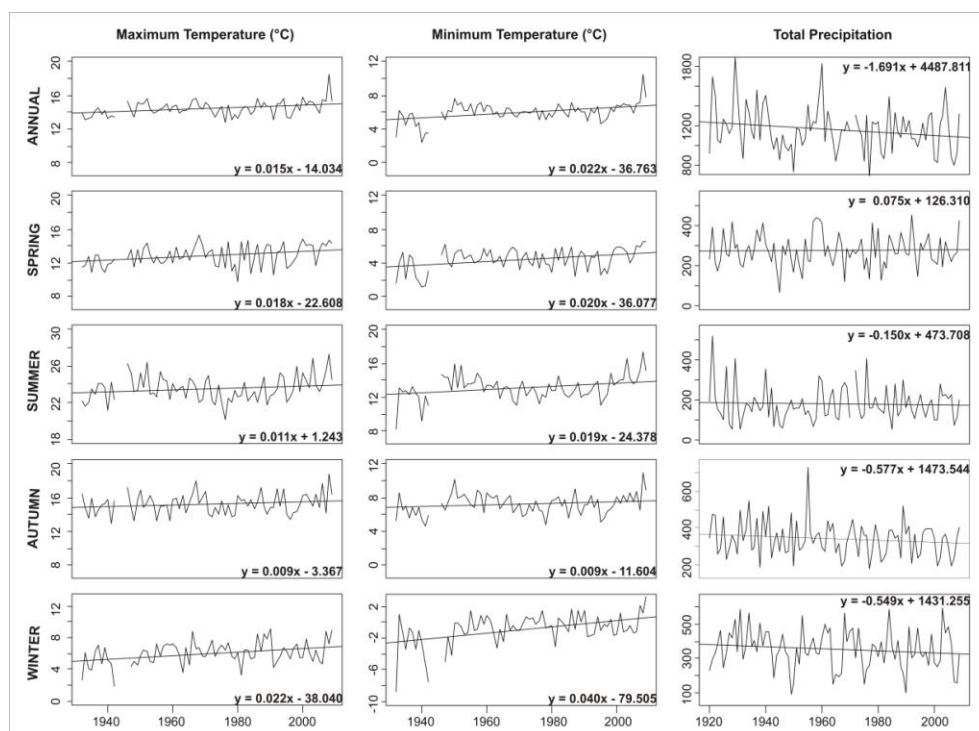


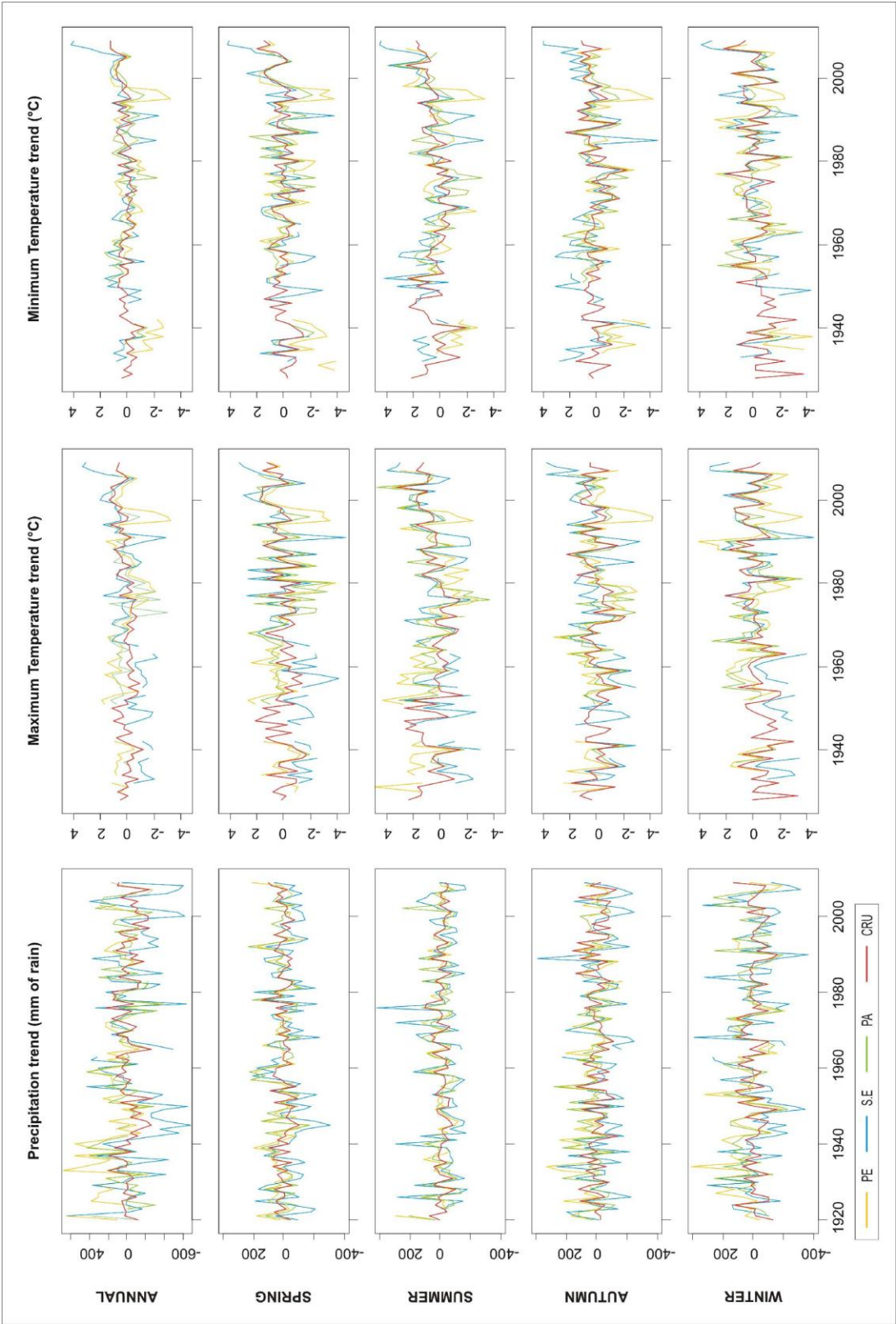
Table A1a. Characteristics of the climatic series for the three meteorological station considered and for the CRU TS 3.1 gridded dataset with 0.5° of spatial resolution (Mitchell and Jones, 2005).

Characteristic of the climatic series				
Meteorological station	Palena	Pescocostanzo	S. Eufemia	CRU TS 3.1
MAXMUM TEMPERATURE				
First year	1936	1928	1932	1901
Last year	2006	2010	2010	2009
Length of the series (years)	71	83	79	108
Missing data (%)	11.3	11.2	8.2	0.0
Mean annual Max. Temperature (°C)	16.35	12.1	15.03	16.53
MINIMUM TEMPERATURE				
First year	1936	1928	1932	1901
Last year	2006	2010	2010	2009
Length of the series (years)	71	83	79	108
Missing data (%)	11.3	11.2	8.2	0.0
Mean annual Min. Temperature (°C)	7.68	3.88	6.40	7.86
PRECIPITATION				
First year	1920	1920	1920	1901
Last year	2008	2011	2011	2009
Length of the series	89	92	92	108
Missing data (%)	1.1	4.2	1.9	0.0
Mean annual Total Precipitation (mm)	992.3	993.7	1437.6	757.8

Table A1b. Pearson's correlation coefficients between climatic series, for the three meteorological station considered and for the CRU TS 3.1 gridded dataset with 0.5° of spatial resolution (Mitchell and Jones, 2005). Coefficients were calculated among annual e seasonal trend (Spring: March-May; Summer: June-August; Autumn: September-November; Winter: December_(t-1)- February). Darker grey highlight significant ($p < 0.001$) correlations between meteorological station and gridded dataset CRU.

Intercorrelation among series											
MAXIMUM TEMPERATURE				MINIMUM TEMPERATURE				PRECIPITATION			
Annual	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>	Annual	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>	Annual	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>
<i>S.Euf.</i>	-0.140			<i>S.Euf.</i>	0.43	1.00		<i>S.Euf.</i>	0.584		
<i>Palena</i>	0.538	0.208		<i>Palena</i>	0.48	0.44	1.00	<i>Palena</i>	0.689	0.613	
<i>CRU</i>	0.208	0.577	0.502	<i>CRU</i>	0.51	0.67	0.61	<i>CRU</i>	0.404	0.464	0.371
Spring	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>	Spring	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>	Spring	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>
<i>S.Euf.</i>	0.293			<i>S.Euf.</i>	0.53	1.00		<i>S.Euf.</i>	0.608		
<i>Palena</i>	0.689	0.534		<i>Palena</i>	0.61	0.48	1.00	<i>Palena</i>	0.812	0.602	
<i>CRU</i>	0.520	0.658	0.705	<i>CRU</i>	0.68	0.64	0.65	<i>CRU</i>	0.483	0.484	0.527
Summer	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>	Summer	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>	Summer	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>
<i>S.Euf.</i>	0.348			<i>S.Euf.</i>	0.43	1.00		<i>S.Euf.</i>	0.791		
<i>Palena</i>	0.594	0.452		<i>Palena</i>	0.57	0.58	1.00	<i>Palena</i>	0.733	0.713	
<i>CRU</i>	0.532	0.645	0.714	<i>CRU</i>	0.46	0.67	0.72	<i>CRU</i>	0.584	0.611	0.572
Autumn	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>	Autumn	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>	Autumn	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>
<i>S.Euf.</i>	0.261			<i>S.Euf.</i>	0.36	1.00		<i>S.Euf.</i>	0.526		
<i>Palena</i>	0.678	0.414		<i>Palena</i>	0.66	0.44	1.00	<i>Palena</i>	0.735	0.647	
<i>CRU</i>	0.671	0.505	0.722	<i>CRU</i>	0.51	0.51	0.74	<i>CRU</i>	0.481	0.399	0.477
Winter	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>	Winter	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>	Winter	<i>Pesco.</i>	<i>S.Euf.</i>	<i>Palena</i>
<i>S.Euf.</i>	0.373			<i>S.Euf.</i>	0.68	1.00		<i>S.Euf.</i>	0.633		
<i>Palena</i>	0.640	0.629		<i>Palena</i>	0.61	0.61	1.00	<i>Palena</i>	0.699	0.610	
<i>CRU</i>	0.517	0.812	0.660	<i>CRU</i>	0.67	0.74	0.77	<i>CRU</i>	0.543	0.562	0.584

Fig. A1c. Annual and seasonal trend of anomalies (w.r.t. 1960-1990) for the three meteorological station considered and for the CRU TS 3.1 gridded dataset with 0.5° of spatial resolution (Mitchell and Jones, 2005). PE: Pescocostanzo; S.E: Sant'Eufemia; PA: Palena; CRU: Gridded dataset.



ANNEX II

Table A2 Critical dates of wood formation and duration for the 10 mountain pine trees (standardized data were averaged per DOY) in CW and OW. Critical dates are indicated as the beginning of enlarging phase (bE), beginning of wall thickening phase (bWT), beginning of mature phase (bM); the cessation of the enlarging phase (cE), cessation of the wall thickening phase (cWT), cessation of the mature phase (cM). Critical duration is indicated as the duration of the enlarging phase (dE = cE-bE), duration of the wall thickening phase (dWT = cWT-bWT) and total duration of the xylogenesis (dX = cWT-bE). Associated standard deviation is indicated as .sd label.

CW																
Tree	bE	bE.sd	bWT	bWT.sd	bM	bM.sd	cE	cE.sd	cWT	cWT.sd	dE	dE.sd	dWT	dWT.sd	dX	dX.sd
1	136	0.7	149	0.6	171	0.5	236	0.9	269	0.7	100	1.1	120	0.9	133	1
2	136	0.7	149	0.6	171	0.5	231	1.7	276	0.6	95	1.8	127	0.8	140	0.9
3	136	0.7	136	0.7	171	0.5	236	0.9	269	0.7	100	1.1	133	1	133	1
4	136	0.7	149	0.6	171	0.5	236	0.9	271	12.3	100	1.1	122	12.3	135	12.3
5	136	0.7	149	0.6	171	0.5	236	0.9	276	0.6	100	1.1	127	0.8	140	0.9
6	136	0.7	136	0.7	171	0.5	255	0.6	276	0.6	119	0.9	140	0.9	140	0.9
7	136	0.7	147	11.6	165	0.6	241	1.8	266	13.9	105	1.9	119	18.1	130	13.9
8	136	0.7	149	0.6	165	0.6	246	0.9	282	0.6	110	1.1	133	0.8	146	0.9
9	136	0.7	149	0.6	171	0.5	246	0.9	276	0.6	110	1.1	127	0.8	140	0.9
10	136	0.7	149	0.6	171	0.5	255	0.6	282	0.6	119	0.9	133	0.8	146	0.9
OW																
Tree	bE	bE.sd	bWT	bWT.sd	bM	bM.sd	cE	cE.sd	cWT	cWT.sd	dE	dE.sd	dWT	dWT.sd	dX	dX.sd
1	136	0.7	149	0.5	171	0.5	236	0.9	265	1.3	100	1.1	116	1.4	129	1.5
2	136	0.7	149	0.5	171	0.5	226	0.8	249	14.3	90	1.1	100	14.3	113	14.3
3	136	0.7	149	0.5	171	0.5	236	0.9	248	19.1	100	1.1	99	19.1	112	19.1
4	136	0.7	149	0.5	171	0.5	236	0.9	249	14.3	100	1.1	100	14.3	113	14.3
5	136	0.7	143	0.5	171	0.5	236	0.9	251	1.3	100	1.1	108	1.4	115	1.5
6	136	0.7	136	0.7	171	0.5	236	0.9	259	19.1	100	1.1	123	19.1	123	19.1
7	136	0.7	149	0.5	171	0.5	236	0.9	258	1.3	100	1.1	109	1.4	122	1.5
8	136	0.7	149	0.6	171	0.5	236	0.9	269	0.7	100	1.1	120	0.9	133	1
9	136	0.7	157	0.8	171	0.5	246	0.9	261	0.6	110	1.1	104	1	125	0.9
10	136	0.7	157	0.8	171	0.5	236	0.9	269	0.7	100	1.1	112	1.1	133	1

Note: Critical dates as well as their associated standard deviations were computed using logistic regressions; the main date corresponds to the 50% probability of the phase being active while the confidence interval limits correspond to the 2.5 and 97.5% probabilities. The standard deviation was obtained by dividing the 95% confidence interval by 2 (Rathgeber 2012).

Fig. A2a Wood formation calendar represented by critical dates (left) and duration (right) of phenological phases in CW (above) and OW (below). Onset and cessation of xylem development phases (enlarging, E; wall thickening, WT; mature, M) are represented by diamond-crossed-by-a-line marks. The left end of the line represents the minimum, the left end of the diamond the first quartile, the middle of the diamond the median, the right end of the diamond the third quartile and the right end of the line the maximum (Rathgeber 2012). Duration of xylem development phases (enlarging, E; wall thickening, WT; total xylogenesis, X) are represented by horizontal bar.

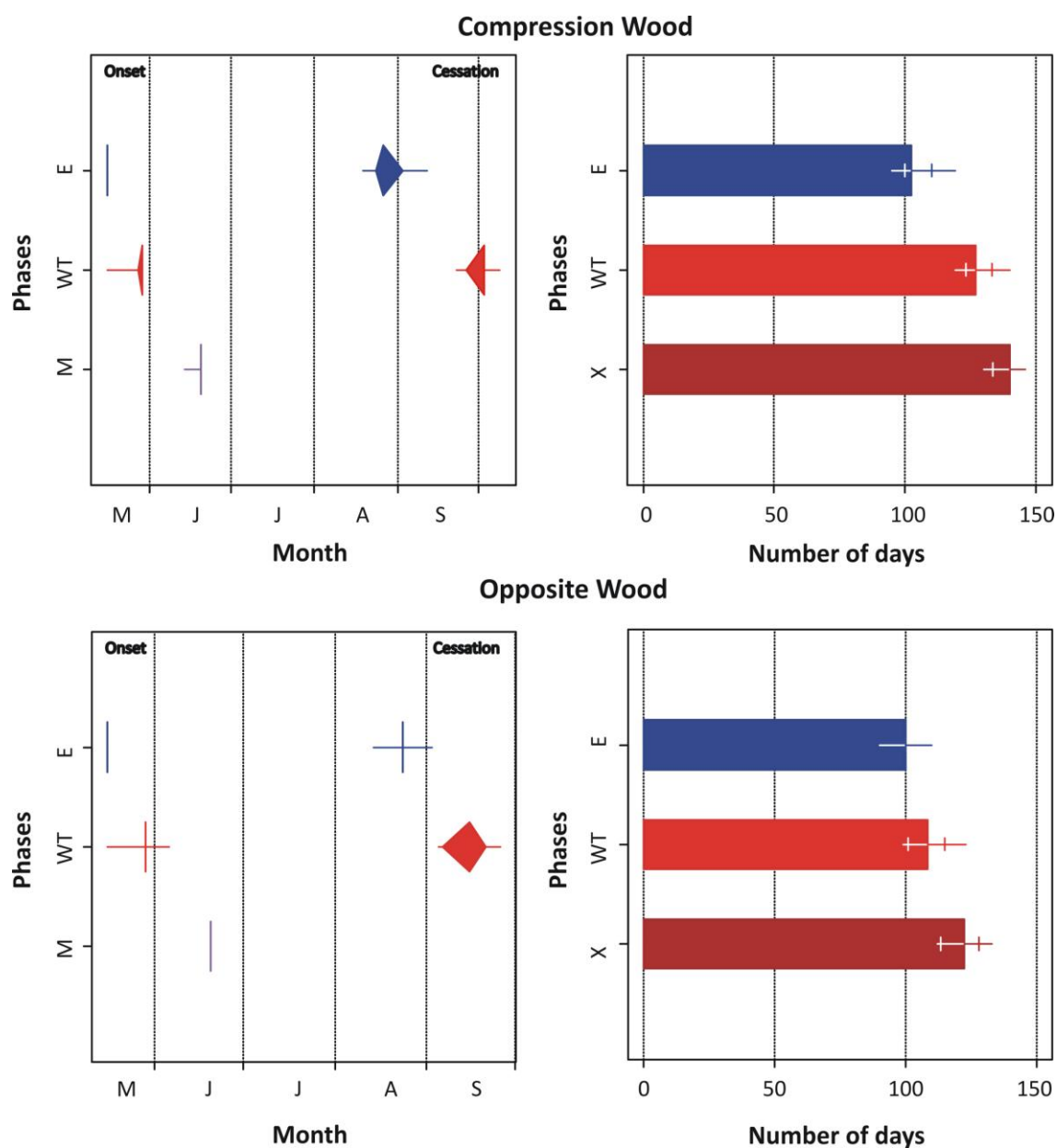
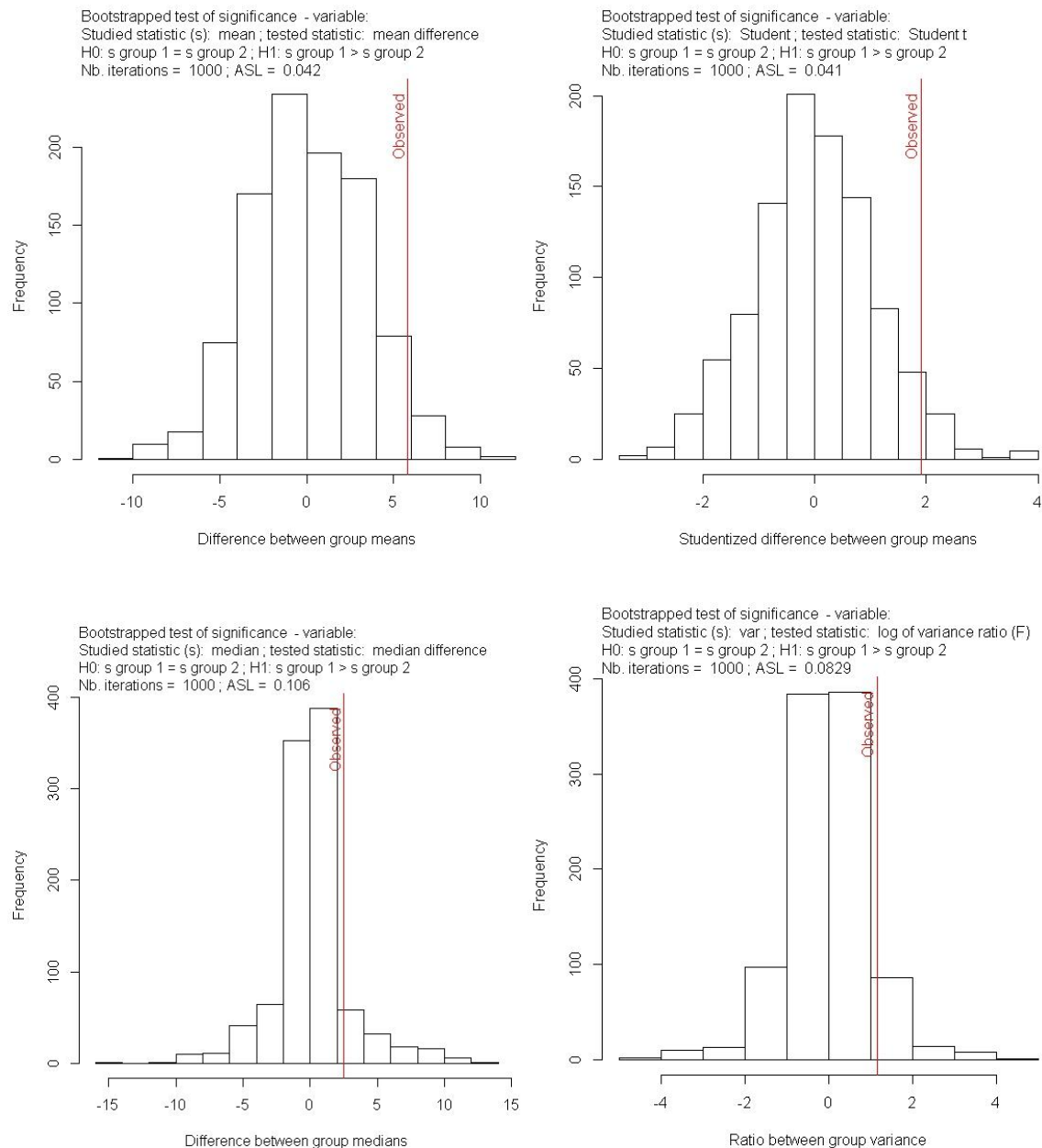
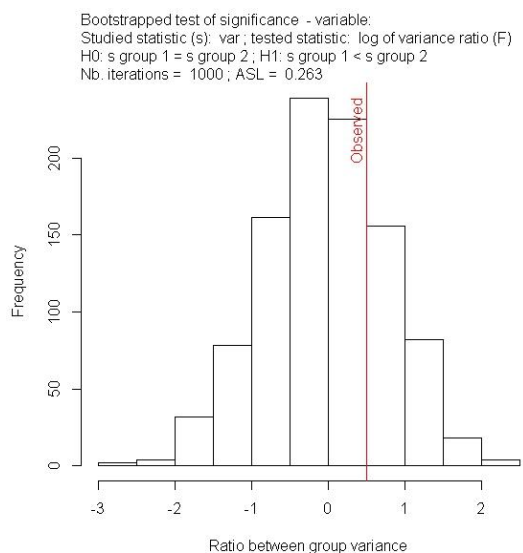
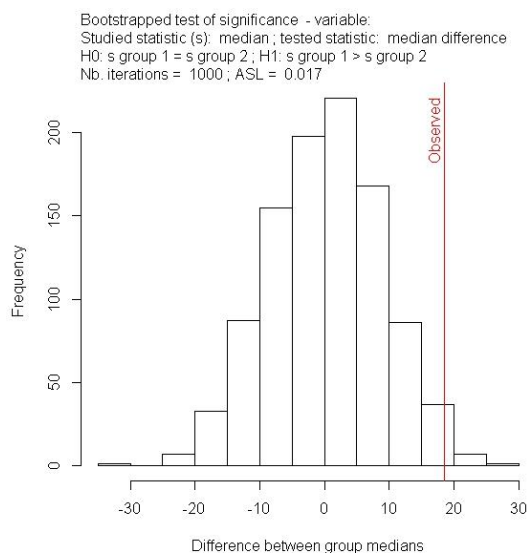
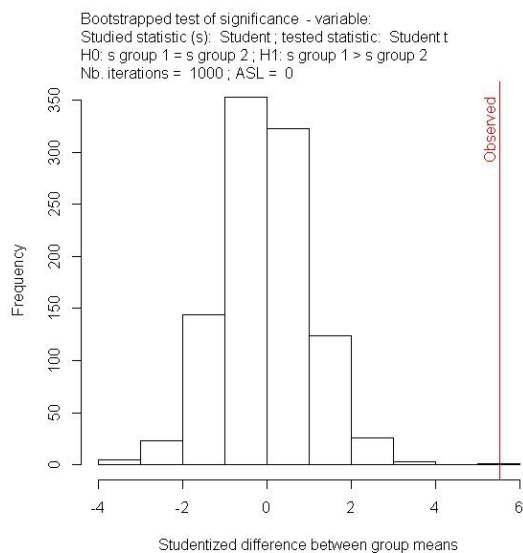
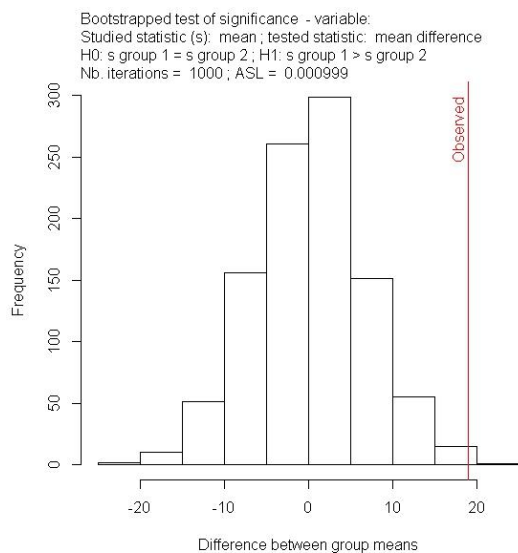


Fig. A2b Results of unilateral bootstrap permutation tests between critical duration of enlarging phase (dE), wall thickening phase (dWT) and total xylogenesis (dX) of CW (group 1) and OW (group 2). Statistic tests: mean differences, Studentized mean difference, median difference and logarithm ratio of variance.

dE)



dWT)



dx)

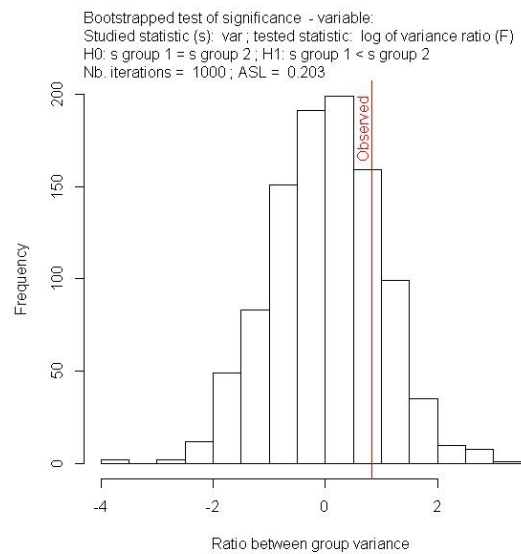
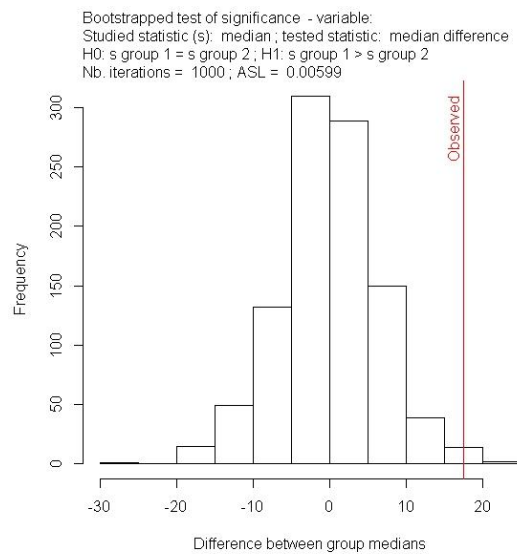
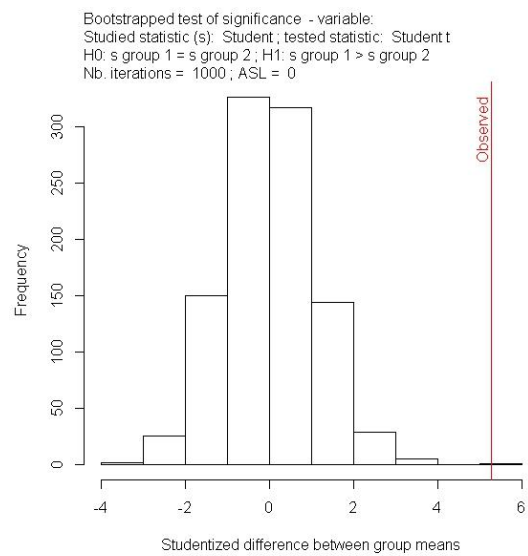
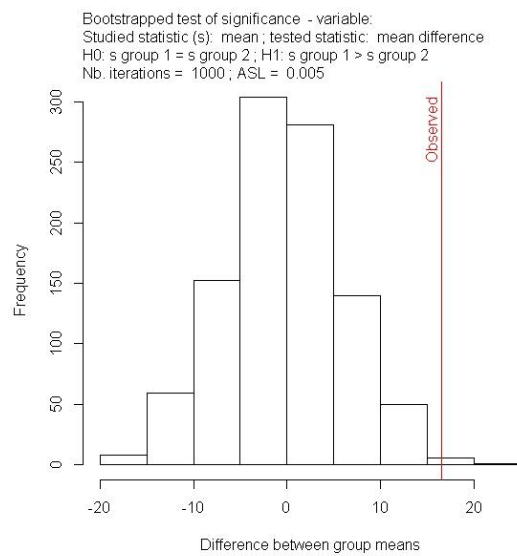
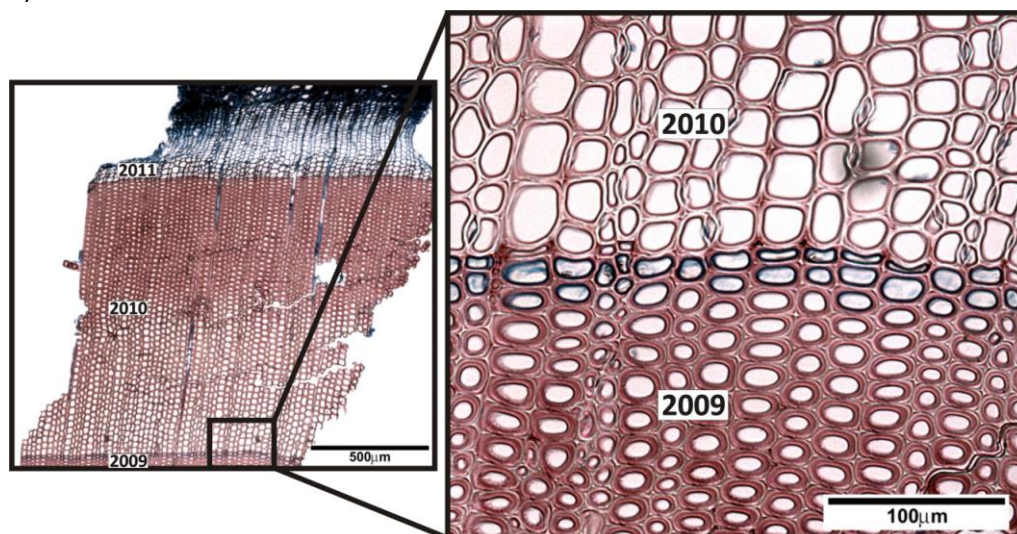


Fig. A2c Uncompleted lignification of the last latewood of CW during the year 2009. Samples taken on: (a) 1 June 2011; (b) 10 August 2011.

a)



b)

